Alinity m

HR HPV AMP Kit

en
HR HPV
REF 09N15-095
53-608175/R02

Revised September 2025

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53-608175/R02
NOTE: Changes Highlighted

Rx ONLY
IVD

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Abbott

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NOTE: Changes Highlighted

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CUSTOMER SERVICE INTERNATIONAL: CALL YOUR ABBOTT REPRESENTATIVE

Instructions must be carefully followed. Reliability of assay results cannot be guaranteed if there are any deviations from these instructions.

NAME

Alinity m HR HPV Assay

- Alinity m HR HPV AMP Kit (List No. 09N15-095)
- Alinity m HR HPV CTRL Kit (List No. 09N15-085)

INTENDED USE

Alinity m HR HPV is a qualitative in vitro test for the detection of Human Papillomavirus DNA. Cervical specimens should be collected by a health care professional using an endocervical collection brush/spatula placed in ThinPrep® PreservCyt® Solution or endocervical broom placed in SurePath™ Preservative Fluid.

Self-collected vaginal specimens, obtained in a healthcare setting, can be tested as an alternative specimen type when cervical sampling is either contraindicated or cervical specimens otherwise cannot be obtained.

This test identifies high-risk (HR) HPV types 16, 18, 45, while reporting the concurrent detection of the other HR genotypes (31/33/52/58) and (35/39/51/56/59/66/68).

Alinity m HR HPV is indicated for use in routine cervical cancer screening as per professional medical guidelines, including triage of ASC-US cytology, co-testing (adjunctive screening) with cytology, and HPV primary screening of women to assess the risk for cervical pre-cancer and cancer. Patients should be followed-up in accordance with professional medical guidelines, results from prior screening, medical history, and other risk factors.

WARNING

If a cervical specimen cannot be obtained, self-collected vaginal specimens should be obtained in a healthcare setting where specimens can be processed by trained personnel and transported to a testing laboratory under controlled conditions. Recommendations for clinical management after testing of either clinician-collected cervical specimens or self-collected vaginal specimens should follow professional guidelines and may differ for these two specimen types.

Alinity m HR HPV is NOT intended:

- for use in determining the need for treatment (ie, excisional or ablative treatment of the cervix) in the absence of high-grade cervical dysplasia. Patients who are HPV 16, 18, or 45 positive should be monitored carefully for the development of high-grade cervical dysplasia according to current practice guidelines.
- for women who have undergone hysterectomy with removal of the cervix.
- for use with samples other than those collected by a clinician using an endocervical brush/spatula placed in ThinPrep PreservCyt Solution or a cervical broom placed in SurePath Preservative Fluid.
- for use with self-collected vaginal specimens other than those collected with collection devices specifically FDA-approved or cleared for use with the Alinity m HR HPV assay.

HPV-negative cancers of the cervix do occur in rare circumstances.[1,2] Also, no cancer screening test is 100% sensitive. Use of this device for primary cervical cancer screening should be undertaken after carefully considering the performance characteristics put forth in this label, as well as recommendations of professional guidelines.

The use of this test has not been evaluated for the management of women with prior ablative or excisional therapy, hysterectomy, who are pregnant or who have other risk factors (eg, HIV+, immunocompromised, history of sexually transmitted infections).

SUMMARY AND EXPLANATION OF THE TEST

Human Papillomavirus (HPV) is a small, non-enveloped, double-stranded DNA virus (approximately 8,000 base pairs) that replicates in the nucleus of squamous epithelial cells and induces hyperproliferative lesions.[3] HPV infections are among the most common sexually transmitted infections.[4] Most HPV infections have a benign clinical consequence and are cleared spontaneously.[5] However, persistent HPV infection in women's cervix may result in progression to cervical cancer.[6-10] More than two-hundred different HPV genotypes have been identified,[11] among which over forty infect mucosal and genital epithelia.[12] Genital HPV genotypes are generally classified into high risk (HR) and low risk (LR) groups based on their carcinogenic potential. HR HPV genotypes (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) are associated with invasive cervical cancer (squamous cell carcinoma or adenocarcinoma) or its immediate precursor (high-grade squamous intraepithelial lesion, cervical intraepithelial neoplasia, carcinoma in situ, or adenocarcinoma in situ), whereas LR HPV genotypes induce benign lesions and are not associated with cervical cancer.[13-16] Three of the 14 HR HPV genotypes, 16, 18, and 45, are associated with approximately 70-80% of invasive cervical cancer cases worldwide.[17-19] For adenocarcinoma, which is more difficult to detect, HPV 16, 18, and 45 have been observed in 78-94% of cases.[17,18,20] Infection by HPV 16 or HPV 18 is associated with higher risk of disease progression compared to other HR HPV genotypes.[21] HPV 66 was categorized as "possibly carcinogenic" based on its relatively low prevalence in invasive cervical carcinomas.[22]

Compared with cervical screening methods identifying cytological abnormalities, molecular tests that specifically detect the presence of HR HPV DNA in cervical cells can potentially increase sensitivity and cost-effectiveness of cervical cancer screening programs.[23-28] Furthermore, HPV DNA tests can be effectively used in triaging patients with equivocal cytology, in post therapeutic follow-up, and in monitoring vaccine efficacy.[29-31]

The Alinity m HR HPV assay is a qualitative in vitro test that amplifies and detects HR HPV DNA in cervical specimens and self-collected vaginal specimens.

The detection of 14 HR HPV genotypes (HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) is achieved through a primer mix targeting a conserved region of the HPV genomes and single stranded DNA probes. The assay can differentiate between HPV 16, HPV 18, HPV 45, and non-HPV 16/18/45 genotypes 31/33/52/58 (Group A, Other HR HPV A) and 35/39/51/56/59/66/68 (Group B, Other HR HPV B).

PRINCIPLES OF THE PROCEDURE

The Alinity m HR HPV assay requires 2 separate assay specific kits:

- Alinity m HR HPV AMP Kit (List No. 09N15-095) consisting of 2 types of multi-well assay trays. The amplification trays (AMP Trays) contain lyophilized, unit-dose PCR amplification/detection reagents. The activation trays (ACT Trays) contain liquid unit-dose activation reagent. The intended storage condition for the Alinity m HR HPV AMP Kit is 2°C to 8°C.
- Alinity m HR HPV CTRL Kit (List No. 09N15-085) consisting of negative controls and positive controls, each supplied as liquid in single-use tubes. The intended storage condition for the Alinity m HR HPV CTRL Kit is −25°C to −15°C.

The Alinity m HR HPV assay utilizes real-time polymerase chain reaction (PCR) to amplify and detect DNA sequences from 14 HR HPV genotypes (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) and human genomic DNA sequences that have been extracted from cervical specimens collected in ThinPrep or SurePath medium, or self-collected vaginal specimens collected with the simpli-COLLECT™ HPV Collection Kit (List No. 09R16-001) or Evalyn® Brush (Rovers® Medical Devices, List No. 09R25-001).

Cervical specimens collected in ThinPrep or SurePath medium are transferred to a transport tube for processing on the Alinity m System. Self-collected vaginal specimens collected with the simpli-COLLECT HPV Collection Kit are resuspended with the Alinity m Specimen Buffer Kit II

(List No. 09R03-050) for processing on the Alinity m System. For self-collected vaginal specimens collected with the Evalyn Brush, the Evalyn Brush head is transferred to an Alinity m Specimen Buffer Kit III tube (List No. 09R03-060) for processing on the Alinity m System.

The steps of the Alinity m HR HPV assay consist of sample preparation, PCR assembly, amplification/detection, and result calculation and reporting. All steps of the Alinity m HR HPV assay procedure are executed automatically by the Alinity m System.

The Alinity m System is designed to be a random access analyzer that can perform the Alinity m HR HPV assay in parallel with other Alinity m assays on the same instrument.

Nucleic acids from specimens are extracted using the Alinity m Sample Prep Kit 1, Alinity m Lysis Solution, Alinity m Diluent Solution, and Alinity m Ethanol Solution or Alinity m Bottle for Ethanol Use (filled with customer supplied 190 Proof Ethanol, ACS, Denaturant free). The Alinity m System employs magnetic microparticle technology to facilitate nucleic acid capture, wash, and elution.

The resulting purified nucleic acids are then combined with liquid unit-dose Alinity m HR HPV activation reagent and lyophilized unit-dose Alinity m HR HPV amplification/detection reagents and transferred into a reaction vessel. Alinity m Vapor Barrier Solution is added to the reaction vessel which is then transferred to an amplification/detection unit for PCR amplification, and real-time fluorescence detection of HR HPV.

The Alinity m HR HPV amplification/detection reagents include primers and probes that amplify and detect all 14 HR HPV genotypes. The assay is designed to report the detection of HPV 16, HPV 18, and HPV 45 as genotype specific results. In addition, the assay reports the detection of the remaining 11 HR HPV genotypes in two groups. Genotype specific probes detect HPV 31, 33, 52, and 58 in one unique fluorescence channel, while genotype specific probes detect HPV 35, 39, 51, 56, 59, 66, and 68 in another unique fluorescence channel.

The Alinity m HR HPV amplification/detection reagents include primers and a probe that amplify and detect an endogenous human beta globin (BG) sequence as a sample validity control for cell adequacy, sample extraction, and amplification efficiency. The Alinity m HR HPV amplification/detection reagent also contains Uracil-DNA Glycosylase (UDG) as a contamination control for amplicons containing uracil, which may be present in molecular laboratories.

Assay controls are tested at or above an established minimum frequency to help ensure that instrument and reagent performance remains satisfactory. During each control event, a negative control and a positive control are processed through sample preparation and PCR procedures that are identical to those used for specimens.

The possibility of nucleic acid contamination on the Alinity m System is minimized because:

- Aerosol barrier pipette tips are used for all pipetting. The pipette tips are discarded after use.
- PCR amplification and detection is carried out automatically in a sealed reaction vessel.
- Disposal of the reaction vessel is performed automatically by the Alinity m System.

For additional information on system and assay technology, refer to the Alinity m System Operations Manual, Section 3.

REAGENTS

Kit Contents

Alinity m HR HPV AMP Kit List No. 09N15-095

The Alinity m HR HPV AMP Kit is comprised of 2 types of multi-well trays: Alinity m HR HPV AMP TRAY 1 and Alinity m HR HPV ACT TRAY 2.

Each Alinity m HR HPV AMP TRAY 1 (individually packed in a foil pouch with a desiccant bag) contains 96 unit-dose lyophilized amplification reagent wells. One reagent well is used per test.

- Amplification reagent wells consist of synthetic oligonucleotides, DNA Polymerase, excipient, dNTPs, and Uracil-DNA Glycosylase in a buffered solution.

Each Alinity m HR HPV ACT TRAY 2 (individually packed in a foil pouch without a desiccant bag) contains 96 unit-dose liquid activation reagent wells. One reagent well is used per test.

- Activation reagent wells consist of magnesium chloride, potassium chloride, and tetramethylammonium chloride. Preservative: 0.15% ProClin® 950.

	Quantity
Σ	384 tests
Alinity m HR HPV AMP TRAY 1	4 trays / 96 tests each
Alinity m HR HPV ACT TRAY 2	4 trays / 96 tests each

WARNINGS AND PRECAUTIONS

IVD

- **For In Vitro Diagnostic Use**

Safety Precautions

Human specimens should be handled as if infectious using safe laboratory procedures, such as those outlined in Biosafety in Microbiological and Biomedical Laboratories,[32] OSHA Standard on Bloodborne Pathogens,[33] CLSI Document M29-A4,[34] and other appropriate biosafety practices.[35] Therefore all human sourced materials should be considered infectious.

These precautions include, but are not limited to, the following:

- Wear gloves when handling specimens or reagents.
- Do not pipette by mouth.
- Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in areas where these materials are handled.
- Clean and disinfect spills of specimens by including the use of a tuberculocidal disinfectant such as 1.0% sodium hypochlorite or other suitable disinfectant.[35]

Decontaminate and dispose of all potentially infectious materials in accordance with local, state and federal regulations.[32]

The following warnings and precautions apply to:
Alinity m HR HPV ACT TRAY 2.

DANGER	Contains Tetramethylammonium chloride, and Methylisothiazolone
H302	Harmful if swallowed.
H316	Causes mild skin irritation[a]
H317	May cause an allergic skin reaction.
H370	Causes damage to organs.
H412	Harmful to aquatic life with long lasting effects.
Prevention	
P260	Do not breathe mist / vapors / spray.
P264	Wash hands thoroughly after handling.
P272	Contaminated work clothing should not be allowed out of the workplace.
P273	Avoid release to the environment.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P301+P312	IF SWALLOWED: Call a POISON CENTER / doctor if you feel unwell.
P302+P352	IF ON SKIN: Wash with plenty of water.
P308+P311	IF exposed or concerned: Call a POISON CENTER / doctor.
P333+P313	If skin irritation or rash occurs: Get medical advice / attention.
P362+P364	Take off contaminated clothing and wash it before reuse.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

[a] Not applicable where regulation EC 1272/2008 (CLP) or OSHA Hazard Communication 29 CFR 1910.1200 (HCS) 2012 have been implemented.

Important information regarding the safe handling, transport, and disposal of this product is contained in the Safety Data Sheet.

Safety Data Sheets are available from your Abbott Representative.

For a detailed discussion of safety precautions during system operation, refer to the Alinity m System Operations Manual, Section 7 and Section 8.

Reagent Shipment

	Shipment Condition
Alinity m HR HPV AMP Kit	On dry ice

Reagent Storage

In order to minimize damage to foil pouches, it is recommended that the Alinity m HR HPV AMP TRAY 1 (AMP TRAY 1) and Alinity m HR HPV ACT TRAY 2 (ACT TRAY 2) are stored in the original kit packaging. Open the foil pouch for the reagent trays just prior to loading onto the instrument. Onboard storage time begins when reagents are loaded on the Alinity m System.

Alinity m HR HPV AMP Kit

	Storage Temperature	Maximum Storage Time
Unopened	2°C to 8°C	Until expiration date
Onboard	System Temperature	30 days (not to exceed expiration date)

Alinity m HR HPV CTRL Kit[a]

	Storage Temperature	Maximum Storage Time
Unopened	−25°C to −15°C	Until expiration date
Onboard	System Temperature	Discard after 4 hours

[a] For detailed information on Alinity m HR HPV CTRL Kit handling and indications of reagent deterioration, refer to the product specific package insert.

Reagent Handling

- Do not use reagents that have been damaged.
- Minimize contact with the surface of reagent trays during handling.
- Up to 2 sets of assay trays (AMP TRAY 1 and ACT TRAY 2) can be loaded on each Alinity m Assay Tray Carrier. AMP Kit lots can be different between the 2 sets of assay trays. However, AMP TRAY 1 and ACT TRAY 2 within each set must be from the same AMP Kit lot.
- The Alinity m System will track the onboard storage time of AMP TRAY 1 and ACT TRAY 2 while on the instrument. The Alinity m System will not allow the use of AMP TRAY 1 and ACT TRAY 2 if the maximum onboard storage time has been exceeded.
- For a detailed discussion of reagent handling precautions during system operation, refer to the Alinity m System Operations Manual, Section 8.

Indications of Reagent Deterioration

- Deterioration of the reagents may be indicated when a control error occurs or control values are repeatedly out of the specified ranges.
- Reagents are shipped on dry ice and are stored at 2°C to 8°C upon arrival. If reagents arrive in a condition contrary to this recommendation or are damaged, immediately contact your Abbott Representative.
- For troubleshooting information, refer to the Alinity m System Operations Manual, Section 10.

INSTRUMENT PROCEDURE

The Alinity m HR HPV assay application specification file must be installed on the Alinity m System prior to performing the assay.

For a detailed description of system operating instructions, refer to the Alinity m System Operations Manual, Section 5.

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Collection

Users must follow the respective manufacturer's instructions for collecting cervical specimens in ThinPrep PreservCyt Solution (Hologic Inc.) or SurePath Preservative Fluid (BD).

Patients must follow the instructions in the simpli-COLLECT HPV Collection Kit (List No. 09R16-001) or Evalyn Brush (List No. 09R25-001) for self-collecting vaginal specimens in a clinical setting.

For vaginal specimens self-collected with the simpli-COLLECT HPV Collection Kit, only the orange-shaft swab provided in the kit should be used. A specimen containing no swab, a different type of swab, or multiple swabs cannot be used with the Alinity m HR HPV assay, and a new specimen should be collected.

Specimen Types

Specimens collected using an endocervical brush/spatula and placed in ThinPrep PreservCyt Solution or using a cervical broom-like collection device and placed in SurePath Preservative Fluid can be used with the Alinity m HR HPV assay. For ThinPrep and SurePath specimens, the aliquot that is removed either prior to or after cytological processing from the collection vial can be used. The aliquot is transferred to Alinity m Transport Tube (List No. 09N49-010 or 09N49-011) or Alinity m Labeled Tube with Pierceable Cap (List No. 09N65-050) and capped with Alinity m Pierceable Cap provided separately (List No. 09N49-012) or together with the tube (List No. 09N49-010 or 09N49-011) for storage.

Self-collected vaginal specimens collected in a clinical setting using the simpli-COLLECT HPV Collection Kit (List No. 09R16-001) or the Evalyn Brush (List No. 09R25-001) can be used with the Alinity m HR HPV assay. Vaginal specimens self-collected using the simpli-COLLECT HPV Collection Kit are prepared using the Alinity m Specimen Buffer Kit II (List No. 09R03-050). For vaginal specimens self-collected using the Evalyn Brush, the Evalyn Brush head is transferred to an Alinity m Specimen Buffer Kit III tube (List No. 09R03-060).

Alinity m HR HPV assay performance with other collection devices or specimen types has not been evaluated.

Collection Device/Medium	Specimen Type[a]
ThinPrep PreservCyt Solution	Cervical Specimens
SurePath Preservative Fluid	Cervical Specimens
simpli-COLLECT HPV Collection Kit	Vaginal Specimens
Evalyn Brush (List No. 09R25-001)	Vaginal Specimens

[a] The instrument does not provide the capability to verify specimen types. It is the responsibility of the operator to use the correct specimen types in the assay.

Specimen Storage

Collection Device/ Medium	Temperature	Maximum Storage Time[a]
ThinPrep PreservCyt Solution	15°C to 30°C 2°C to 8°C −25°C to −15°C	6 Months 6 Months 6 Months
SurePath Preservative Fluid	15°C to 30°C 2°C to 8°C −25°C to −15°C	14 Days 3 Months 3 Months
simpli-COLLECT HPV Collection Kit	2°C to 8°C 15°C to 30°C 2°C to 30°C −25°C to −15°C	14 days (dry)[f] 8 days (dry)[f] 14 days (resuspended)[b,c] 14 days (resuspended)[b,c,d]
Evalyn Brush	2°C to 30°C 2°C to 30°C −25°C to −15°C	14 days (dry)[g] 14 days (resuspended)[b,e] 14 days (resuspended)[b,d,e]

[a] From date of collection.
[b] Total storage time for dry and resuspended specimens must not exceed a combined total of 14 days from date of collection.
[c] Resuspend the specimen in Alinity m Specimen Buffer II.
[d] Avoid more than 4 freeze-thaw cycles.
[e] Resuspend the specimen in an Alinity m Specimen Buffer III tube pre-filled with buffer.
[f] Vaginal specimens collected using simpli-COLLECT HPV may be shipped (up to 7 days) and then stored at 2°C to 8°C for up to a total (including shipping time) of 14 days or at 15°C to 30°C for up to a total (including shipping time) of 8 days before resuspension in specimen buffer.
[g] Vaginal specimens collected using the Evalyn Brush may be shipped (up to 7 days) and then stored at 2°C to 30°C for up to a total (including shipping time) of 14 days before resuspension in specimen buffer.

Specimen Shipping

Ship specimens according to the recommended storage temperature and time listed in the **Specimen Storage** section. Package and label specimens in compliance with applicable state, federal, and international regulations covering the transport of clinical, diagnostic, or biological specimens.

Preparation for Analysis

Cervical Specimens

ThinPrep specimens and SurePath specimens need to be transferred to an Alinity m Transport Tube (List No. 09N49-010 or 09N49-011) or Alinity m Labeled Tube with Pierceable Cap (List No. 09N65-050) for processing on the Alinity m System. Vortex each specimen for 15 to 20 seconds. Ensure that the contents in the ThinPrep or SurePath vial are on the bottom by tapping the vial on the bench.

Immediately transfer a volume of each specimen based on aliquot source to a transport tube prior to loading on the Alinity m System.

Specimen Type	Aliquot Source	Transport Tube Specimen Volume[c]
ThinPrep	Pre-Cytology Specimen[a]	0.55 mL – 2.0 mL
	Post-Cytology Specimen	
	Specimen Not Intended for Cytology	
SurePath	Pre-Cytology Specimen[b]	0.5 mL (single aliquot)
	Post-Cytology Specimen	0.5 mL – 2.0 mL
	Specimen Not Intended for Cytology	

[a] No more than a single aliquot may be removed from the ThinPrep specimen prior to cytology processing. Refer to manufacturer's instructions for use.

[b] No more than a single aliquot (no more than 0.5 mL) can be removed from the SurePath specimen prior to cytology processing. Refer to manufacturer's instructions for use.

[c] Transport tube specimen volume includes 0.4 mL instrument aspiration volume and dead volume.

The transport tubes may be capped with an Alinity m Pierceable Cap (List No. 09N49-012). When handling specimens in transport tubes, do not touch the top of pierceable caps to avoid contamination. All specimen tubes must be labeled with specimen ID barcodes, or must be identified with a specimen ID and rack and position.

Note: Do not use specimens which appear bloody or have a dark brown color.

Note: Do not use Alinity m Aliquot Tube (List No. 09N49-013). Refer to the Assay Procedure section for sample instructions prior to loading on the Alinity m System.

Self-Collected Vaginal Specimens

Self-collected vaginal specimens using the simpli-COLLECT HPV Collection Kit:

1. Uncap the Alinity m Sample Tube for Dry Swab and place in a sample rack or a tube holder.
2. Transfer 2.5 mL of Alinity m Specimen Buffer from the Alinity m Specimen Buffer Kit II (List No. 09R03-050) into the Alinity m Sample Tube for Dry Swab.
3. Replace the pierceable cap on the tube.

Note: The swab should remain in the tube while being processed on the Alinity m System.

Self-collected vaginal specimens using the Evalyn Brush (List No. 09R25-001):

1. Uncap an Alinity m Specimen Buffer Kit III tube (List No. 09R03-060). Place uncapped tube in a sample rack or a tube holder to avoid spilling buffer out of the tube; place the pierceable cap on a clean table liner to avoid contamination.
2. Hold the Evalyn Brush device and remove the pink cap by squeezing the cap sides.
3. Push the pink plunger to expose the brush head and the base of the brush head. Do not touch the Evalyn Brush white bristles.
4. If using Evalyn Tweezers to detach the brush head, position the tweezers around the base of the brush head. If using a ReMover to transfer the brush head, guide the Evalyn Brush into the ReMover until the casing hits the ReMover.

Note: A new pair of Evalyn Tweezers or a new ReMover must be used for each brush.

5. Squeeze the Evalyn Tweezers or the ReMover firmly together and pull the brush head off. Keep the Evalyn Tweezers or ReMover compressed.
6. Hold the Evalyn Tweezers or ReMover over the mouth of the Alinity m Specimen Buffer Kit III tube and release the brush head into the tube with the bristles oriented up.
7. Replace the pierceable cap on the tube.

Refer to the Assay Procedure section for sample instructions prior to loading on the Alinity m System.

PROCEDURE

Materials Provided

09N15-095 Alinity m HR HPV AMP Kit

Materials Required but not Provided

- 09N15-085 Alinity m HR HPV CTRL Kit
- 09N18-001 Alinity m Sample Prep Kit 1
- 09N20-001 Alinity m Lysis Solution
- 09N20-002 Alinity m Ethanol Solution or 09N20-012 Alinity m Bottle for Ethanol Use (filled with customer supplied 190 Proof Ethanol, ACS, Denaturant free)
- 09N20-003 Alinity m Diluent Solution
- 09N20-004 Alinity m Vapor Barrier Solution
- 09N49-010 Alinity m Transport Tube Pierceable Capped
- 09N49-011 Alinity m Transport Tubes
- 09N49-012 Alinity m Pierceable Caps
- 09N65-050 Alinity m Labeled Tube with Pierceable Cap
- 09R03-050 Alinity m Specimen Buffer Kit II
- 09R03-060 Alinity m Specimen Buffer Kit III
- Alinity m HR HPV Application Specification File - 09N15-03B or higher
- Vortex mixer
- Calibrated pipettes capable of delivering 100 µL to 1000 µL
- Aerosol barrier pipette tips for 100 µL to 1000 µL pipettes
- Plate adapter for 384 well plates (eg, Eppendorf Catalog No. 022638955)
- Centrifuge with swing plate rotor capable of accommodating the plate adapter and capable of ≥ 100 *g*

Use the simpli-COLLECT HPV Collection Kit (List No. 09R16-001) or Evalyn Brush (List No. 09R25-001) for self-collected vaginal specimens.

For information on materials required for operation of the instrument, refer to the Alinity m System Operations Manual, Section 1.

For general operating procedures, refer to the Alinity m System Operations Manual, Section 5.

For optimal performance, it is important to perform routine maintenance as described in the Alinity m System Operations Manual, Section 9.

Procedural Precautions

- Read the instructions in this package insert carefully before processing samples.
- When handling specimens in transport tubes, do not touch the top of pierceable caps to avoid cross-contamination. Specimens can contain high levels of organisms. Change gloves if they come in contact with specimen.
- Use aerosol barrier pipette tips or disposable pipettes only one time when pipetting specimens. To prevent contamination to the pipette barrel while pipetting, care should be taken to avoid touching the pipette barrel to the inside of the sample tube or container. The use of extended aerosol barrier pipette tips is recommended.
- Do not use specimens if the specimen tube is damaged or if buffer has leaked from the specimen tube. Discard unused, damaged or leaking specimen tubes in accordance with local, state, and federal regulations.
- Self-collected vaginal specimens collected using the simpli-COLLECT HPV Collection Kit must be resuspended with Alinity m Specimen Buffer from Alinity m Specimen Buffer Kit II prior to loading on the Alinity m System. Do not remove swab from the Alinity m Transport Tube.
- Self-collected vaginal specimens collected using the Evalyn Brush must be resuspended in the Alinity m Specimen Buffer Kit III tube prior to loading on the Alinity m System. Do not remove the Evalyn Brush head from the tube.
- Work area and instrument platforms must be considered potential sources of contamination.
- Ensure the Alinity m HR HPV AMP TRAY 1 is tapped prior to loading on the Alinity m System per instructions in the **Assay Procedure** section.
- Ensure the Alinity m HR HPV ACT TRAY 2 is centrifuged prior to loading on the Alinity m System per instructions in the **Assay Procedure** section.
- Monitoring procedures for the presence of amplification product can be found in the Alinity m System Operations Manual, Section 9.
- To reduce the risk of nucleic acid contamination, clean and disinfect spills of specimens by including the use of a tuberculocidal disinfectant such as 1.0% sodium hypochlorite or other suitable disinfectant.

- To prevent contamination, change to new gloves before handling the Alinity m Sample Prep Kit 1, assay trays, system solutions, Integrated Reaction Unit (IRU) sleeves, and pipette tips. Also change to new gloves whenever they are contaminated by a specimen, a control, or a reagent. Always use powder-free gloves.
- The use of the Alinity m HR HPV CTRL Kit is integral to the performance of the Alinity m HR HPV assay. Refer to the **QUALITY CONTROL PROCEDURES** section of this package insert for details. Refer to the Alinity m HR HPV CTRL Kit package insert for preparation and usage.
- The Alinity m HR HPV control reagents are contained in single-use tubes with pierceable caps. Avoid contamination or damage to the caps after removal from their original packaging. Discard tubes after use.

Assay Procedure

Prior to loading on the Alinity m System, hold the AMP TRAY 1 by the edges with the label facing up and tap 3 times on the bench. Carefully transfer the AMP TRAY 1 to the Alinity m Assay Tray Carriers.

Prior to loading on the Alinity m System, ACT TRAY 2 must be centrifuged as follows:

1. Load ACT TRAY 2 onto the plate adapter (eg, Eppendorf Catalog No. 022638955).
2. Load the plate adapter (with ACT TRAY 2) on a swing plate centrifuge capable of accommodating the plate adapter. Spin at 100 *g* to 800 *g* for 1 to 5 minutes to remove potential bubbles.
3. Immediately following centrifugation, carefully transfer the ACT TRAY 2 to the Alinity m Assay Tray Carriers. Take care to minimize disturbance to the ACT TRAY 2. Load the tray carriers per the Alinity m System Operations Manual, Section 5.
4. If disturbance occurs during transfer that could potentially introduce bubbles (eg, dropping, bumping, inversion of the ACT TRAY 2), re-centrifuge the ACT TRAY 2.
5. Proceed with the **Reagent and sample inventory management** procedure per the Alinity m System Operations Manual, Section 5.

For a detailed description of how to run an assay, refer to the Alinity m System Operations Manual, Section 5.

Prior to testing specimens, check control status. If control testing is required, refer to **QUALITY CONTROL PROCEDURES** section. Controls may be tested separately or with specimens.

For preparation of samples, refer to the instructions under **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS**: **Preparation for Analysis** section. If the specimen is stored frozen, it must be completely thawed prior to sample preparation.

Vortex specimens in transport tubes for 15 to 20 seconds prior to loading on the Alinity m System. From the Specimen tab on the Create Order screen, select the appropriate assay (HR HPV) being tested.

The Alinity m System will track the onboard storage time of amplification reagents, controls, and specimens while on the instrument. The Alinity m System will not allow the use of amplification reagents, controls, or process specimens that have exceeded the allowable onboard storage time. Specimen tubes need to meet the requirements for minimum sample volume (refer to table below) and use of caps when loaded on the Alinity m System.

Specimen and Alinity m HR HPV CTRL tubes may be placed on the Alinity m Universal Sample Rack (sample rack) onboard the system for up to 4 hours. The Alinity m HR HPV AMP Kits may be stored onboard the Alinity m System for up to 30 days.

Vaginal specimens collected with the simpli-COLLECT HPV Collection Kit and Evalyn Brush must be loaded onto the sample rack capped and with the retention bar on. Clean the retention bar after each use.

Tube Type[a, b]	List No.	Specimen Volume (Minimum - Maximum)	Cap Requirement on Instrument
Alinity m Transport Tube Pierceable Capped	09N49-010	ThinPrep: 0.55 mL – 2.00 mL SurePath: 0.50 mL – 2.00 mL	Capped or Uncapped
Alinity m Transport Tube Alinity m Pierceable Cap	09N49-011 09N49-012	ThinPrep: 0.55 mL – 2.00 mL SurePath: 0.50 mL – 2.00 mL	Capped or Uncapped
Alinity m Labeled Tube with Pierceable Cap	09N65-050	ThinPrep: 0.55 mL – 2.00 mL SurePath: 0.50 mL – 2.00 mL	Capped or Uncapped
Alinity m Sample Tube for Dry Swab	AB887	Fill volume allows initial test and retests down to 0.55 mL	Capped
Alinity m Specimen Buffer Kit III (for Evalyn Brush)	09R03-060	Fill volume allows initial test and one retest	Capped

[a] The Transport Tube is the same commodity in all configurations.

[b] Do not use Alinity m Aliquot Tube (List No. 09N49-013)

Post Processing Procedure

Upon completion of Alinity m sample preparation, ThinPrep and SurePath cervical specimens that have been transferred to an Alinity m Transport Tube, self-collected vaginal specimens collected with the simpli-COLLECT HPV Collection Kit and resuspended in Alinity m Specimen Buffer from Alinity m Specimen Buffer Kit II, and self-collected vaginal specimens collected with the Evalyn Brush and transferred into an Alinity m Specimen Buffer Kit III tube can be recapped using new, unused Alinity m Pierceable Caps (List No. 09N49-012). When recapping, take care not to touch the top of new pierceable caps to avoid cross-contamination. Change gloves if they come in contact with specimen liquid. Store specimens according to the **Specimen Storage** section.

QUALITY CONTROL PROCEDURES

Negative and Positive Controls

One Alinity m HR HPV Negative CTRL and one Alinity m HR HPV Positive CTRL are recommended to be tested, at or above the minimum frequency of once every 24 hours, to monitor the performance of the assay and Alinity m System. Valid results for all control levels must be obtained before specimen results are reported. Additional controls may be tested in accordance with local, state, and/or federal regulations or accreditation requirements and your laboratory's quality control policy.

If quality control results do not meet the acceptance criteria, refer to the Alinity m System Operations Manual, Section 10, for troubleshooting information. A flag is displayed for specimens when a control result is invalid. All of the specimens processed following an invalid assay control must be retested as volume permits (refer to minimum volume requirements in the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section).

If control results are invalid, refer to the Alinity m System Operations Manual, Section 5 for a description of quality control flags, and Section 10 for troubleshooting information. HPV must not be positive in the negative control. HPV positivity in the negative control is indicative of contamination by other samples or by amplified product. To avoid contamination, clean the Alinity m System and repeat sample processing for controls and specimens following the Procedural Precautions in this package insert.

Monitoring procedures for the presence of amplification product can be found in the Alinity m System Operations Manual, Section 9.

If negative controls are persistently positive, contact your Abbott Representative.

Detection of Inhibition and/or Cell Inadequacy

The Alinity m HR HPV assay detects an endogenous human beta globin (BG) sequence as a Cellular Control (CC) signal to evaluate cell adequacy, sample extraction, and amplification efficiency. The CC cycle number (CN) is assessed against a defined range to determine specimen validity.

A Flag or Message Code is displayed when CC CN value of a specimen is out of the established range.

- If the HPV signal(s) in a specimen is positive and CC CN is out of range, the specimen will yield an HR HPV Positive interpretation. A Flag will be displayed.
- If the HPV signal(s) in a specimen is negative and CC CN is out of range, no HPV result will be reported and a Message Code will be displayed.

Refer to the Alinity m System Operations Manual, Section 5 for an explanation of the corrective actions for Flags.

Refer to the Alinity m System Operations Manual, Section 10 for an explanation of the corrective actions for Message Codes.

RESULTS

Results and interpretations for the Alinity m HR HPV assay are determined based on the comparison of specimen's cycle number (CN) values in each HPV signal against signal-specific, established cutoff values. Five HPV signals measured in separate fluorescence channels corresponding to HPV 16, HPV 18, HPV 45, HPV 31/33/52/58 (pooled

signal: a single signal corresponding to any individual genotype or combination of genotypes within the group) and HPV 35/39/51/56/59/66/68 (pooled signal) are evaluated for each sample. Each signal is either determined as “Positive” if the CN is less than or equal to a fixed assay cutoff cycle for that signal or is determined as “Negative” if either the CN is not generated or the CN is greater than the assay cutoff cycle. All positive signals (HPV 16, HPV 18, HPV 45, Other HR HPV A, or Other HR HPV B) are reported in the sample result. Other HR HPV A is reported when HPV 31/33/52/58 signal is positive. Other HR HPV B is reported when HPV 35/39/51/56/59/66/68 signal is positive. Samples with any positive HR HPV signals have an interpretation of “HR HPV Positive.” Samples with all negative HR HPV signals will have an interpretation of “Negative.”

Interpretation of Results

Results	Interpretation
Negative	Negative
HPV16	HR HPV Positive (16 Positive)
HPV18	HR HPV Positive (18 Positive)
HPV45	HR HPV Positive (45 Positive)
Other HR HPV A	HR HPV Positive (A Positive)
Other HR HPV B	HR HPV Positive (B Positive)
HPV16; HPV18	HR HPV Positive (16;18 Positive)
HPV16; HPV45	HR HPV Positive (16;45 Positive)
HPV16; Other HR HPV A	HR HPV Positive (16;A Positive)
HPV16; Other HR HPV B	HR HPV Positive (16;B Positive)
HPV18; HPV45	HR HPV Positive (18;45 Positive)
HPV18; Other HR HPV A	HR HPV Positive (18;A Positive)
HPV18; Other HR HPV B	HR HPV Positive (18;B Positive)
HPV45; Other HR HPV A	HR HPV Positive (45;A Positive)
HPV45; Other HR HPV B	HR HPV Positive (45;B Positive)
Other HR HPV A; Other HR HPV B	HR HPV Positive (A;B Positive)
HPV16; HPV18; HPV45	HR HPV Positive (16;18;45 Positive)
HPV16; HPV18; Other HR HPV A	HR HPV Positive (16;18;A Positive)
HPV16; HPV18; Other HR HPV B	HR HPV Positive (16;18;B Positive)
HPV16; HPV45; Other HR HPV A	HR HPV Positive (16;45;A Positive)
HPV16; HPV45; Other HR HPV B	HR HPV Positive (16;45;B Positive)
HPV16; Other HR HPV A; Other HR HPV B	HR HPV Positive (16;A;B Positive)
HPV18; HPV45; Other HR HPV A	HR HPV Positive (18;45;A Positive)
HPV18; HPV45; Other HR HPV B	HR HPV Positive (18;45;B Positive)
HPV18; Other HR HPV A; Other HR HPV B	HR HPV Positive (18;A;B Positive)
HPV45; Other HR HPV A; Other HR HPV B	HR HPV Positive (45;A;B Positive)
HPV16; HPV18; HPV45; Other HR HPV A	HR HPV Positive (16;18;45;A Positive)
HPV16; HPV18; HPV45; Other HR HPV B	HR HPV Positive (16;18;45;B Positive)
HPV16; HPV18; Other HR HPV A; Other HR HPV B	HR HPV Positive (16;18;A;B Positive)
HPV16; HPV45; Other HR HPV A; Other HR HPV B	HR HPV Positive (16;45;A;B Positive)
HPV18; HPV45; Other HR HPV A; Other HR HPV B	HR HPV Positive (18;45;A;B Positive)
HPV16; HPV18; HPV45; Other HR HPV A; Other HR HPV B	HR HPV Positive (16;18;45;A;B Positive)

Flags, Results Codes, and Message Codes

Some results may contain information in the Flags and Codes fields. For a description of the flags and result codes that may appear in these fields, refer to the Alinity m System Operations Manual, Section 5.

For a description of message codes, refer to the Alinity m System Operations Manual, Section 10.

LIMITATIONS OF THE PROCEDURE

- Alinity m HR HPV assay performance has been evaluated for use with the Alinity m HR HPV AMP Kit, Alinity m HR HPV CTRL Kit, Alinity m Sample Prep Kit 1, Alinity m Lysis Solution, Alinity m Ethanol Solution (or customer-supplied 190 proof Ethanol ACS), Alinity m Diluent Solution, Alinity m Vapor Barrier Solution, Alinity m Transport Tube Pierceable Capped (Alinity m Pierceable Caps may be supplied separately), Alinity m Transport Tubes, Alinity m Labeled Tube with Pierceable Cap, Alinity m HR HPV Application Specification File, and the Alinity m System.
- Optimal performance of this test requires appropriate specimen collection and handling (refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert).
- Alinity m HR HPV has been validated for use with cervical specimens collected by a health care professional using an endocervical brush/spatula and placed in ThinPrep PreservCyt Solution or using a cervical broom-like collection device and placed in SurePath Preservative Fluid. Alinity m HR HPV assay has also been validated for use with self-collected vaginal specimens collected with the simpli-COLLECT HPV Collection Kit or Evalyn Brush in a clinical setting. Use only validated self-collection devices for vaginal swab specimens.
- The use of this test has not been evaluated for the management of women with prior ablative or excisional therapy, hysterectomy, who are pregnant, or who have other risk factors (eg, HIV positive, immunocompromised, history of sexually transmitted infections).
- Alinity m HR HPV has not been evaluated in cases of suspected abuse and for other medico-legal indications.
- The instruments and assay procedures reduce the risk of contamination by amplification product. However, nucleic acid contamination from the positive controls or specimens must be controlled by good laboratory practice and careful adherence to the procedures specified in this package insert.
- If the HPV results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.
- As with any diagnostic test, results from the Alinity m HR HPV assay should be interpreted in conjunction with other clinical and laboratory findings.

- Clinical performance was evaluated in women with an intact cervix who were aged 25-85 years old.
- The performance of Alinity m HR HPV with ThinPrep PreservCyt Solution and SurePath Preservative Fluid has not been adequately established for HPV vaccinated individuals; performance was evaluated in a limited population of vaccinated individuals.
- Alinity m HR HPV was not evaluated in women less than 25 years old with ASC-US cytology results.
- Detection of an Other HR HPV A (HPV 31/33/52/58) genotype at low level in SurePath specimens was inhibited by the presence of high concentrations (10^5 Copies/mL) of HPV 18. Detection of an Other HR HPV B (HPV 35/39/51/56/59/66/68) genotype at low level in SurePath specimens was inhibited by the presence of high concentrations (10^5 Copies/mL) of HPV 16, 18, or 45. No inhibition was observed for Other HR HPV A or Other HR HPV B when competing targets were at 10^4 Copies/mL.
- Assay interference may be observed for specimens collected in ThinPrep PreservCyt Solution in the presence of Monistat-1 at concentrations greater than 0.75%. Assay interference may also be observed for vaginal specimens in the presence of mucus at concentrations greater than 2.5%.
- Based on reported CN values, detection of HPV may be impacted for cervical specimens stored in SurePath in the presence of mucus at a concentration of 5% w/v.
- Cross-contamination of samples can cause false positive results. Carryover was observed at the Sample Input/Preparation stage 0.83% (4/481) with a 95% CI: 0.32% to 2.12% on the Alinity m System.
- The percent of invalid Alinity m HR HPV test results in ThinPrep was 0.06% (7/11,528) with 95% CI: 0.03% to 0.13%, in SurePath was 1.14% (109/9,575) with a 95% CI: 0.94% to 1.37%, in simpli-COLLECT specimens was 0.4% (7/1,768) with a 95% CI: 0.2% to 0.8%, and in Evalyn Brush specimens was 0.8% (14/1,756) with a 95% CI: 0.5% to 1.3%.
- Infection with HPV is not an indicator of cytologic HSIL or underlying high-grade CIN, nor does it imply that CIN2/3 or cancer will develop. Most women infected with one or more high-risk HPV types do not develop CIN2/3 or cancer. Women who are positive for high-risk HPV by the Alinity m HR HPV assay from a clinician-collected cervical specimen or a self-collected vaginal specimen should follow-up with a clinician to determine management.
- A negative high-risk HPV result does not exclude the possibility of future cytologic HSIL or underlying CIN2/3 or cancer.
- This test does not detect DNA of HPV low-risk types (eg, 6, 11, 42, 43, 44) since there is no clinical utility for testing of low-risk HPV types.[36]
- Detection of high-risk HPV is dependent on the number of virus copies present in the specimen and may be affected by specimen collection methods, patient factors, stage of infection, and the presence of interfering substances.
- Prevalence of HPV infection in a population may affect performance. Positive predictive values decrease when testing populations with low prevalence or individuals with no risk of infection.
- Human β-globin amplification and detection is included in Alinity m HR HPV to differentiate HPV negative specimens from those that do not exhibit HPV signal due to insufficient cell mass in the specimen. All HPV negative specimens must have a valid β-globin signal within a pre-defined range to be identified as valid negatives.
- Use of this product must be limited to personnel trained on the Alinity m HR HPV assay and the use of the Alinity m System.
- The effects of other potential variables such as vaginal discharge, use of tampons, douching, etc. and specimen collection variables have not been evaluated.
- Though rare, mutations within the highly conserved regions of the genomic DNA of Human papillomavirus covered by Alinity m HR HPV primers and/or probes may result in failure to detect the presence of the viral DNA.
- The presence of PCR inhibitors may cause false negative or invalid results.
- HPV Negative results are not intended to prevent women from proceeding to colposcopy.
- Positive test results indicate the presence of any one or more of the high-risk types, but since patients may be co-infected with low-risk types it does not rule out the presence of low-risk types in patients with mixed infections.
- Residual ThinPrep PreservCyt post-cytology specimens evaluated were processed on the ThinPrep 2000 and ThinPrep 5000 Processors. Residual SurePath Preservative Fluid post-cytology specimens evaluated were processed on the BD PrepMate™/ PrepStain™ Processor.

SPECIFIC PERFORMANCE CHARACTERISTICS

Limit of Detection at the Clinical Cutoff

Alinity m HR HPV assay limit of detection (LoD) at the clinical cutoff was determined by testing plasmids for 14 HR HPV genotype sequences (HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) in pooled HR HPV negative clinical specimens collected in ThinPrep PreservCyt Solution (ThinPrep clinical matrix), pooled HR HPV negative clinical specimens collected in SurePath Preservative Fluid (SurePath clinical matrix), and Specimen Buffer containing HR HPV negative C33A cells (contrived vaginal matrix). The LoD was also determined for HPV positive cell lines SiHa (HPV 16) and HeLa (HPV 18) in ThinPrep clinical matrix, SurePath clinical matrix, contrived vaginal matrix, and clinical vaginal matrix. Each plasmid and cell line was diluted at multiple concentrations including below and above the expected LoD level for each matrix. HPV 16, 18, 45, 58 (Other HR HPV A), and 39 (Other HR HPV B) plasmids, and SiHa and HeLa cell lines were tested with 3 lots of amplification reagents, while the remaining HR HPV plasmids (HPV 31, 33, 35, 51, 52, 56, 59, 66, and 68) were tested with 2 lots of amplification reagents. For each lot, a minimum of 20 replicates were tested across 3 days for each plasmid and cell line target concentration in each matrix. The LoD was defined as the lowest target concentration having a ≥95% positivity rate with all higher concentrations having a ≥95% positivity rate. **Table 1** includes results from the concentration near the LoD for the reagent lot with the most conservative LoD for ThinPrep clinical matrix. **Table 2** includes results from the concentration near the LoD for the reagent lot with the most conservative LoD for SurePath clinical matrix. **Table 3** includes results from the concentration near the LoD for the reagent lot with the most conservative LoD for contrived vaginal matrix. The analytical sensitivity for SiHa (HPV16) and HeLa (HPV18) was found to be equivalent between contrived vaginal matrix and clinical vaginal matrix. The concentration in bold indicates the lowest target concentration having a ≥95% positivity rate with all higher concentrations having a ≥95% positivity rate.

Table 1. Limit of Detection for HR HPV Plasmids and SiHa and HeLa Cell Lines in ThinPrep Clinical Matrix

HPV Target	Concentration[a]	No. Valid	No. Positive	Positivity Rate	95% Confidence Interval
SiHa (HPV 16)	80	24	24	100.0%	(86.2%, 100.0%)
	40	**23**	**22**	**95.7%**	**(79.0%, 99.2%)**
	20	24	20	83.3%	(64.1%, 93.3%)
HeLa (HPV 18)	18.8	23	23	100.0%	(85.7%, 100.0%)
	9.2	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	4.8	24	20	83.3%	(64.1%, 93.3%)
HPV 16	960	24	24	100.0%	(86.2%, 100.0%)
	480	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	240	20	18	90.0%	(69.9%, 97.2%)
HPV 18	240	24	24	100.0%	(86.2%, 100.0%)
	120	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	60	24	22	91.7%	(74.2%, 97.7%)
HPV 31	600	24	24	100.0%	(86.2%, 100.0%)
	300	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	150	24	22	91.7%	(74.2%, 97.7%)
HPV 33	1200	24	24	100.0%	(86.2%, 100.0%)
	600	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	300	24	22	91.7%	(74.2%, 97.7%)
HPV 35	600	24	24	100.0%	(86.2%, 100.0%)
	300	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	150	24	22	91.7%	(74.2%, 97.7%)
HPV 39	19200	20	20	100.0%	(83.9%, 100.0%)
	9600	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	4800	24	22	91.7%	(74.2%, 97.7%)
HPV 45	240	24	24	100.0%	(86.2%, 100.0%)
	120	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	60	24	22	91.7%	(74.2%, 97.7%)
HPV 51	1200	22	22	100.0%	(85.1%, 100.0%)
	600	**22**	**21**	**95.5%**	**(78.2%, 99.2%)**
	300	22	16	72.7%	(51.8%, 86.8%)
HPV 52	5000	24	24	100.0%	(86.2%, 100.0%)
	2500	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	1250	24	21	87.5%	(69.0%, 95.7%)
HPV 56	600	24	24	100.0%	(86.2%, 100.0%)
	300	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	150	24	19	79.2%	(59.5%, 90.8%)
HPV 58	2400	24	24	100.0%	(86.2%, 100.0%)
	1200	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	600	24	22	91.7%	(74.2%, 97.7%)
HPV 59	300	20	20	100.0%	(83.9%, 100.0%)
	150	**20**	**20**	**100.0%**	**(83.9%, 100.0%)**
	75	24	21	87.5%	(69.0%, 95.7%)
HPV 66	300	20	20	100.0%	(83.9%, 100.0%)
	150	**20**	**20**	**100.0%**	**(83.9%, 100.0%)**
	75	24	22	91.7%	(74.2%, 97.7%)
HPV 68	600	23	23	100.0%	(85.7%, 100.0%)
	300	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	150	22	3	13.6%	(4.7%, 33.3%)

[a] Copies/assay for plasmids, Cells/assay for SiHa and HeLa cell lines.

Table 2. Limit of Detection for HR HPV Plasmids and SiHa and HeLa Cell Lines in SurePath Clinical Matrix

HPV Target	Concentration[a]	No. Valid	No. Positive	Positivity Rate	95% Confidence Interval
SiHa (HPV 16)	160	23	23	100.0%	(85.7%, 100.0%)
	80	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	40	24	22	91.7%	(74.2%, 97.7%)
HeLa (HPV 18)	9.2	24	24	100.0%	(86.2%, 100.0%)
	4.8	**23**	**22**	**95.7%**	**(79.0%, 99.2%)**
	2.4	22	15	68.2%	(47.3%, 83.6%)
HPV 16	240	24	24	100.0%	(86.2%, 100.0%)
	120	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	60	24	18	75.0%	(55.1%, 88.0%)
HPV 18	240	24	23	95.8%	(79.8%, 99.3%)
	120	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	60	24	22	91.7%	(74.2%, 97.7%)
HPV 31	600	23	23	100.0%	(85.7%, 100.0%)
	300	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	150	24	22	91.7%	(74.2%, 97.7%)
HPV 33	1200	24	24	100.0%	(86.2%, 100.0%)
	600	**23**	**22**	**95.7%**	**(79.0%, 99.2%)**
	300	24	22	91.7%	(74.2%, 97.7%)
HPV 35	1200	24	24	100.0%	(86.2%, 100.0%)
	600	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	300	24	22	91.7%	(74.2%, 97.7%)
HPV 39	2400	24	23	95.8%	(79.8%, 99.3%)
	1200	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	600	22	20	90.9%	(72.2%, 97.5%)
HPV 45	240	24	24	100.0%	(86.2%, 100.0%)
	120	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	60	24	22	91.7%	(74.2%, 97.7%)
HPV 51	1200	24	24	100.0%	(86.2%, 100.0%)
	600	**23**	**22**	**95.7%**	**(79.0%, 99.2%)**
	300	22	20	90.9%	(72.2%, 97.5%)
HPV 52	2500	24	24	100.0%	(86.2%, 100.0%)
	1250	**24**	**24**	**100.0%**	**(86.2%, 100.0%)**
	625	24	22	91.7%	(74.2%, 97.7%)
HPV 56	600	24	24	100.0%	(86.2%, 100.0%)
	300	**24**	**23**	**95.8%**	**(79.8%, 99.3%)**
	150	22	20	90.9%	(72.2%, 97.5%)
HPV 58	600	20	20	100.0%	(83.9%, 100.0%)
	300	**20**	**20**	**100.0%**	**(83.9%, 100.0%)**
	150	24	20	83.3%	(64.1%, 93.3%)
HPV 59	2400	23	22	95.7%	(79.0%, 99.2%)
	1200	**23**	**23**	**100.0%**	**(85.7%, 100.0%)**
	600	24	20	83.3%	(64.1%, 93.3%)
HPV 66	4800	24	24	100.0%	(86.2%, 100.0%)
	2400	**22**	**22**	**100.0%**	**(85.1%, 100.0%)**
	1200	21	18	85.7%	(65.4%, 95.0%)
HPV 68	9600	20	20	100.0%	(83.9%, 100.0%)
	4800	**20**	**20**	**100.0%**	**(83.9%, 100.0%)**
	2400	20	16	80.0%	(58.4%, 91.9%)

[a] Copies/assay for plasmids, Cells/assay for SiHa and HeLa cell lines.

Table 3. Limit of Detection for HR HPV Plasmids and SiHa and HeLa Cell Lines in Contrived Vaginal Matrix

HPV Target	Concentration[a]	No. Valid	No. Positive	Positivity Rate	95% Confidence Interval
SiHa (HPV 16)	20	24	24	100.0	(86.2%, 100.0%)
	10	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	5	24	21	87.5	(69.0%, 95.7%)
HeLa (HPV 18)	4.6	22	22	100.0	(85.1%, 100.0%)
	2.3	**22**	**21**	**95.5**	**(78.2%, 99.2%)**
	1.2	21	17	81.0	(60.0%, 92.3%)
HPV 16	240	24	24	100.0	(86.2%, 100.0%)
	120	**24**	**23**	**95.8**	**(79.8%, 99.3%)**
	60	23	7	30.4	(15.6%, 50.9%)
HPV 18	30	24	24	100.0	(86.2%, 100.0%)
	15	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	7.5	24	10	41.7	(24.5%, 61.2%)
HPV 31	150	24	24	100.0	(86.2%, 100.0%)
	75	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	37.5	24	16	66.7	(46.7%, 82.0%)
HPV 33	300	24	24	100.0	(86.2%, 100.0%)
	150	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	75	24	15	62.5	(42.7%, 78.8%)
HPV 35	240	24	24	100.0	(86.2%, 100.0%)
	120	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	60	24	14	58.3	(38.8%, 75.5%)
HPV 39	600	24	24	100.0	(86.2%, 100.0%)
	300	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	150	24	22	91.7	(74.2%, 97.7%)
HPV 45	60	24	24	100.0	(86.2%, 100.0%)
	30	**24**	**23**	**95.8**	**(79.8%, 99.3%)**
	15	24	11	45.8	(27.9%, 64.9%)
HPV 51	1200	24	24	100.0	(86.2%, 100.0%)
	600	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	300	24	7	29.2	(14.9%, 49.2%)
HPV 52	1250	24	24	100.0	(86.2%, 100.0%)
	625	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	312.5	24	22	91.7	(74.2%, 97.7%)
HPV 56	320	24	24	100.0	(86.2%, 100.0%)
	160	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	80	24	16	66.7	(46.7%, 82.0%)
HPV 58	600	24	23	95.8	(79.8%, 99.3%)
	300	**23**	**23**	**100.0**	**(85.7%, 100.0%)**
	150	24	19	79.2	(59.5%, 90.8%)
HPV 59	200	24	24	100.0	(86.2%, 100.0%)
	100	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	50	23	8	34.8	(18.8%, 55.1%)
HPV 66	80	24	24	100.0	(86.2%, 100.0%)
	40	**24**	**23**	**95.8**	**(79.8%, 99.3%)**
	20	24	5	20.8	(9.2%, 40.5%)
HPV 68	2400	24	24	100.0	(86.2%, 100.0%)
	1200	**24**	**24**	**100.0**	**(86.2%, 100.0%)**
	600	24	22	91.7	(74.2%, 97.7%)

a Copies/assay for plasmids, Cells/assay for SiHa and HeLa cell lines.

Precision

Within-Laboratory Precision Study 1

Within-laboratory precision of the Alinity m HR HPV assay was evaluated with a 22-member panel of samples in ThinPrep PreservCyt Solution and a 13-member panel of samples in SurePath Preservative Fluid. The ThinPrep panel included 6 pools made from clinical specimens collected in ThinPrep PreservCyt Solution and 16 contrived samples prepared with SiHa (HPV 16), HeLa (HPV 18), HPV 45, HPV 39, or HPV 58 plasmids in ThinPrep PreservCyt Solution containing HR HPV negative C33A cells. The SurePath panel included 6 pools made from clinical specimens collected in SurePath Preservative Fluid and 7 contrived samples prepared with SiHa or HeLa in SurePath Preservative Fluid containing HR HPV negative C33A cells. For each sample type, 1 negative clinical pool and 5 moderate positive clinical pools representative of each genotype category (HPV 16, 18, 45, Other HR HPV A, and Other HR HPV B) were prepared. ThinPrep contrived panels consisted of high negative, low positive, and moderate positive target levels for each genotype category, in addition to a negative panel member. SurePath contrived panels consisted of high negative, low positive, and moderate positive target levels for SiHa and HeLa cell lines, in addition to a negative panel member. Each panel member was tested with 2 replicates twice each day for 12 days on 3 Alinity m Systems with 3 reagent lots.

A description of the panels, percentages of replicates (positive, negative with virus detected and negative with virus not detected), and analysis of cycle number (CN) variability are presented in **Table 4** and **Table 5** for the ThinPrep panel and in **Table 6** and **Table 7** for the SurePath panel. For ThinPrep panel members, the total %CV for HPV CN ranged from 1.4% to 6.5% across all low positive or higher genotype panel members (**Table 5**). For SurePath panel members, the total %CV for HPV CN ranged from 1.4% to 5.7% across all low positive or higher genotypes panel members (**Table 7**).

Table 4. Summary of Within-Laboratory Precision Study 1 Panel Members, Median CN, and Positivity Rates: ThinPrep

Panel	Panel Description	N[a]	Median CN[b]	% Positive (n/N)[c]	% Negative Virus detected (n/N)	% Negative Virus not detected (n/N)
Pooled negative clinical sample	Negative	144	NA	0.0 (0/144)	0.0 (0/144)	100.0 (144/144)
HPV negative cell line	Negative	144	NA	0.0 (0/144)	0.0 (0/144)	100.0 (144/144)
HPV 16 positive SiHa cell line	HPV 16 high negative	144	32.86	56.9 (82/144)	24.3 (35/144)	18.8 (27/144)
HPV 18 positive HeLa cell line	HPV 18 high negative	144	36.00	10.4 (15/144)	2.8 (4/144)	86.8 (125/144)
HPV 45 plasmid	HPV 45 high negative	144	32.20	37.5 (54/144)	61.1 (88/144)	1.4 (2/144)
Other HR HPV A plasmid[d]	Other HR HPV A high negative	144	36.00	17.4 (25/144)	11.1 (16/144)	71.5 (103/144)
Other HR HPV B plasmid[e]	Other HR HPV B high negative	144	36.00	33.3 (48/144)	9.0 (13/144)	57.6 (83/144)
Pooled clinical sample	HPV 16 moderate positive	144	27.67	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 18 moderate positive	144	28.01	98.6 (142/144)	0.0 (0/144)	1.4 (2/144)
	HPV 45 moderate positive	144	28.37	98.6 (142/144)	0.0 (0/144)	1.4 (2/144)
	Other HR HPV A moderate positive	144	27.72	99.3 (143/144)	0.0 (0/144)	0.7 (1/144)
	Other HR HPV B moderate positive	144	27.00	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
HPV 16 positive SiHa cell line	HPV 16 low positive	144	28.89	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 16 moderate positive	144	27.45	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
HPV 18 positive HeLa cell line	HPV 18 low positive	144	28.29	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 18 moderate positive	144	27.15	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
HPV 45 plasmid	HPV 45 low positive	144	30.50	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 45 moderate positive	144	29.95	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
Other HR HPV A plasmid[d]	Other HR HPV A low positive	144	30.18	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	Other HR HPV A moderate positive	144	29.96	99.3 (143/144)	0.7 (1/144)	0.0 (0/144)
Other HR HPV B plasmid[e]	Other HR HPV B low positive	144	30.73	97.2 (140/144)	0.0 (0/144)	2.8 (4/144)
	Other HR HPV B moderate positive	144	28.75	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)

[a] N is the number of valid replicates.

[b] The median CN value reported was calculated for all replicates, regardless whether the CN was ≤ or > the clinical cutoff. Negative replicates with no virus detected were assigned the last cycle of PCR (36.00). Due to 0% virus detection, median CN was not reported for negative panels (NA).

[c] The % Positive was based on the number of valid replicates with a CN value ≤ the clinical cutoff.

[d] HPV 58 was used for the Other HR HPV A panel.

[e] HPV 39 was used for the Other HR HPV B panel.

Table 5. Variance Component Analysis Results for Within-Laboratory Precision Study 1: ThinPrep

					Within-Run Component		Between-Run Component		Between-Day Component		Between-Instrument/Lot Component		Total[c]	
Panel	Panel Description	N[a]	n[b]	Mean CN	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Pooled clinical sample	HPV 16 moderate positive	144	144	27.52	1.330	4.8	0.747	2.7	0.000	0.0	0.000	0.0	1.525	5.5
	HPV 18 moderate positive	144	142	27.67	1.425	5.1	0.344	1.2	0.362	1.3	0.213	0.8	1.525	5.5[f]
	HPV 45 moderate positive	144	142	28.08	1.084	3.9	0.000	0.0	0.192	0.7	0.080	0.3	1.104	3.9[f]
	Other HR HPV A moderate positive	144	143	27.17	1.510	5.6	0.852	3.1	0.000	0.0	0.310	1.1	1.762	6.5[f]
	Other HR HPV B moderate positive	144	144	27.01	0.771	2.9	0.441	1.6	0.000	0.0	0.000	0.0	0.889	3.3
HPV 16 positive SiHa cell line	HPV 16 low positive	144	144	28.89	0.378	1.3	0.087	0.3	0.175	0.6	0.000	0.0	0.425	1.5
	HPV 16 moderate positive	144	144	27.45	0.382	1.4	0.000	0.0	0.030	0.1	0.101	0.4	0.396	1.4
HPV 18 positive HeLa cell line	HPV 18 low positive	144	144	28.30	0.752	2.7	0.000	0.0	0.205	0.7	0.204	0.7	0.806	2.8
	HPV 18 moderate positive	144	144	27.22	0.570	2.1	0.261	1.0	0.000	0.0	0.213	0.8	0.662	2.4
HPV 45 plasmid	HPV 45 low positive	144	144	30.52	0.373	1.2	0.025	0.1	0.123	0.4	0.196	0.6	0.439	1.4
	HPV 45 moderate positive	144	144	29.96	0.373	1.2	0.000	0.0	0.172	0.6	0.090	0.3	0.421	1.4
Other HR HPV A plasmid[d]	Other HR HPV A low positive	144	144	30.23	0.386	1.3	0.000	0.0	0.133	0.4	0.000	0.0	0.409	1.4
	Other HR HPV A moderate positive	144	143	29.91	0.319	1.1	0.000	0.0	0.193	0.6	0.173	0.6	0.411	1.4[f]
Other HR HPV B plasmid[e]	Other HR HPV B low positive	144	140	30.75	0.369	1.2	0.000	0.0	0.227	0.7	0.205	0.7	0.479	1.6[f]
	Other HR HPV B moderate positive	144	144	28.83	0.327	1.1	0.000	0.0	0.146	0.5	0.167	0.6	0.395	1.4

[a] N is the number of valid replicates.

[b] n is the number of valid positive replicates with a CN value ≤ the clinical cutoff.

[c] Total includes Within-Run, Between-Run, Between-Day, and Between-Instrument/Lot Components.

[d] HPV 58 was used for the Other HR HPV A panel.

[e] HPV 39 was used for the Other HR HPV B panel.

[f] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Table 6. Summary of Within-Laboratory Precision Study 1 Panel Members, Median CN, and Positivity Rates: SurePath

Panel	Panel Description	N[a]	Median CN[b]	% Positive (n/N)[c]	% Negative Virus detected (n/N)	% Negative Virus not detected (n/N)
Pooled negative clinical sample	Negative	144	NA	0.0 (0/144)	0.0 (0/144)	100.0 (144/144)
HPV negative cell line	Negative	144	NA	0.0 (0/144)	0.0 (0/144)	100.0 (144/144)
HPV 16 positive SiHa cell line	HPV 16 high negative	144	36.00	25.0 (36/144)	0.7 (1/144)	74.3 (107/144)
HPV 18 positive HeLa cell line	HPV 18 high negative	144	36.00	14.6 (21/144)	0.0 (0/144	85.4 (123/144)
Pooled clinical sample	HPV 16 moderate positive	144	28.39	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 18 moderate positive	144	27.14	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 45 moderate positive	144	28.24	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	Other HR HPV A moderate positive	144	27.39	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	Other HR HPV B moderate positive	144	28.22	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
HPV 16 positive SiHa cell line	HPV 16 low positive	144	29.35	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
	HPV 16 moderate positive	144	27.91	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)
HPV 18 positive HeLa cell line	HPV 18 low positive	144	28.07	97.2 (140/144)	0.0 (0/144)	2.8 (4/144)
	HPV 18 moderate positive	144	26.77	100.0 (144/144)	0.0 (0/144)	0.0 (0/144)

[a] N is the number of valid replicates.

[b] The median CN value reported was calculated for all replicates, regardless whether the CN was ≤ or > the clinical cutoff. Negative replicates with no virus detected were assigned the last cycle of PCR (36.00). Due to 0% virus detection, median CN was not reported for negative panels (NA).

[c] The % Positive was based on the number of valid replicates with a CN value ≤ the clinical cutoff.

Table 7. Variance Component Analysis Results for Within-Laboratory Precision Study 1: SurePath

					Within-Run Component		Between-Run Component		Between-Day Component		Between-Instrument/Lot Component		Total[c]	
Panel	Panel Description	N[a]	n[b]	Mean CN	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Pooled clinical sample	HPV 16 moderate positive	144	144	28.23	0.743	2.6	0.449	1.6	0.167	0.6	0.140	0.5	0.896	3.2
	HPV 18 moderate positive	144	144	27.21	1.271	4.7	0.000	0.0	0.347	1.3	0.000	0.0	1.318	4.8
	HPV 45 moderate positive	144	144	28.18	0.441	1.6	0.222	0.8	0.158	0.6	0.029	0.1	0.520	1.8
	Other HR HPV A moderate positive	144	144	26.93	1.493	5.5	0.000	0.0	0.330	1.2	0.132	0.5	1.535	5.7
	Other HR HPV B moderate positive	144	144	28.19	0.600	2.1	0.304	1.1	0.212	0.8	0.000	0.0	0.705	2.5
HPV 16 positive SiHa cell line	HPV 16 low positive	144	144	29.38	0.370	1.3	0.092	0.3	0.171	0.6	0.081	0.3	0.425	1.4
	HPV 16 moderate positive	144	144	27.93	0.309	1.1	0.117	0.4	0.160	0.6	0.110	0.4	0.383	1.4
HPV 18 positive HeLa cell line	HPV 18 low positive	144	140	28.26	0.834	3.0	0.000	0.0	0.311	1.1	0.000	0.0	0.890	3.1[d]
	HPV 18 moderate positive	144	144	26.71	0.483	1.8	0.192	0.7	0.000	0.0	0.000	0.0	0.520	1.9

[a] N is the number of valid replicates.

[b] n is the number of valid positive replicates with a CN value ≤ the clinical cutoff.

[c] Total includes Within-Run, Between-Run, Between-Day, and Between-Instrument/Lot Components

[d] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Within-Laboratory Precision Study 2

An additional within-laboratory precision study was conducted with a 6-member panel of contrived samples in SurePath Preservative Fluid. Contrived panel members were prepared with HPV 45, HPV 39, or HPV 58 plasmids in SurePath Preservative Fluid containing HR HPV negative C33A cells. Each HPV genotype was targeted to low positive and moderate positive target levels. Each panel member was tested with a minimum of 2 replicates twice each day for 12 days on each of 3 reagent lots on each of 3 instruments.

A description of the panels, percentages of replicates (positive, negative with virus detected and negative with virus not detected), and analysis of cycle number (CN) variability are presented in **Table 8** and **Table 9** for the SurePath panel. The total %CV for HPV CN ranged from 1.0% to 1.2% across all genotypes targeted at low positive or higher (**Table 9**).

Table 8. Summary of Within-Laboratory Precision Study 2 Panel Members, Median CN, and Positivity Rates: SurePath

Panel	Panel Description	N[a]	Median CN[b]	% Positive (n/N)[c]	% Negative Virus detected (n/N)	% Negative Virus not detected (n/N)
HPV 45 plasmid	HPV 45 low positive	643	29.06	99.8 (642/643)	0.2 (1/643)	0.0 (0/643)
	HPV 45 moderate positive	646	28.90	100.0 (646/646)	0.0 (0/646)	0.0 (0/646)
Other HR HPV A plasmid[d]	Other HR HPV A low positive	647	30.76	97.4 (630/647)	0.3 (2/647)	2.3 (15/647)
	Other HR HPV A moderate positive	645	29.77	99.5 (642/645)	0.2 (1/645)	0.3 (2/645)
Other HR HPV B plasmid[e]	Other HPV B low positive	594	31.40	86.4 (513/594)	5.4 (32/594)	8.2 (49/594)
	Other HPV B moderate positive	647	30.39	99.8 (646/647)	0.0 (0/647)	0.2 (1/647)

[a] N is the number of valid replicates.

[b] The median CN value reported was calculated for all replicates, regardless whether the CN was ≤ or > the clinical cutoff. Negative replicates with no virus detected were assigned the last cycle of PCR (36.00).

[c] The % Positive was based on the number of valid replicates with a CN value ≤ the clinical cutoff.

[d] HPV 58 was used for the Other HR HPV A panel.

[e] HPV 39 was used for the Other HR HPV B panel.

Table 9. Variance Component Analysis Results for Within-Laboratory Precision Study 2: SurePath

Panel	Panel Description	N[a]	n[b]	Mean CN	Within-Run Component SD	Within-Run Component %CV	Between-Run Component SD	Between-Run Component %CV	Between-Day Component SD	Between-Day Component %CV	Between-Lot Component SD	Between-Lot Component %CV	Between-Instrument Component SD	Between-Instrument Component %CV	Total[c] SD	Total[c] %CV
HPV 45 plasmid	HPV 45 low positive	643	642	29.06	0.238	0.8	0.121	0.4	0.000	0.0	0.000	0.0	0.111	0.4	0.289	1.0[f]
	HPV 45 moderate positive	646	646	28.90	0.247	0.9	0.067	0.2	0.061	0.2	0.044	0.2	0.170	0.6	0.316	1.1
Other HR HPV A plasmid[d]	Other HR HPV A low positive	647	630	30.76	0.314	1.0	0.084	0.3	0.000	0.0	0.065	0.2	0.057	0.2	0.336	1.1[f]
	Other HR HPV A moderate positive	645	642	29.78	0.332	1.1	0.000	0.0	0.074	0.2	0.026	0.1	0.107	0.4	0.358	1.2[f]
Other HR HPV B plasmid[e]	Other HR HPV B low positive	594	513	31.33	0.282	0.9	0.137	0.4	0.000	0.0	0.000	0.0	0.062	0.2	0.320	1.0[f]
	Other HR HPV B moderate positive	647	646	30.39	0.306	1.0	0.046	0.2	0.075	0.2	0.098	0.3	0.093	0.3	0.346	1.1[f]

[a] N is the number of valid replicates.
[b] n is the number of valid positive replicates with a CN value ≤ the clinical cutoff.
[c] Total includes Within-Run, Between-Run, Between-Day, Between-Lot, and Between-Instrument Components.
[d] HPV 58 was used for the Other HR HPV A panel.
[e] HPV 39 was used for the Other HR HPV B panel.
[f] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Lot-to-Lot Precision

The variability of Alinity m HR HPV reagent lots was assessed with HPV positive panels in ThinPrep and SurePath matrices. ThinPrep matrix consisted of ThinPrep PreservCyt Solution containing HPV negative C33A cells and SurePath matrix consisted of SurePath Preservative Fluid containing C33A cells. For both matrices, HPV positive panels were prepared with HPV 16, HPV 18, HPV 45, HPV 58, or HPV 39 plasmids targeted to low positive and moderate positive concentrations. Each positive panel member was tested with 3 different reagent lots on 1 Alinity m System. Twenty or twenty-four replicates were tested per panel member per lot. For ThinPrep panel members, the total %CV for HPV CN ranged from 1.1% to 2.1% across all genotypes (**Table 10**). For SurePath panel members, the total %CV for HPV CN ranged from 1.0% to 2.0% across all genotypes (**Table 11**).

Table 10. Lot-to-Lot Precision Study: ThinPrep

Panel	Panel Description	N[b]	n[c]	% Positive	Mean CN	Within-Lot Component SD	Within-Lot Component %CV	Between-Lot Component SD	Between-Lot Component %CV	Total[a] SD	Total[a] %CV
HPV 16 plasmid	Low positive	72	72	100.0	29.94	0.314	1.0	0.097	0.3	0.329	1.1
	Moderate positive	72	72	100.0	28.90	0.301	1.0	0.109	0.4	0.321	1.1
HPV 18 plasmid	Low positive	72	72	100.0	28.87	0.347	1.2	0.190	0.7	0.395	1.4
	Moderate positive	72	72	100.0	27.99	0.310	1.1	0.109	0.4	0.328	1.2
HPV 45 plasmid	Low positive	72	72	100.0	30.08	0.315	1.0	0.108	0.4	0.333	1.1
	Moderate positive	72	72	100.0	29.15	0.300	1.0	0.132	0.5	0.328	1.1
Other HR HPV A plasmid[d]	Low positive	60	60	100.0	30.71	0.344	1.1	0.368	1.2	0.504	1.6
	Moderate positive	60	60	100.0	29.11	0.274	0.9	0.220	0.8	0.351	1.2
Other HR HPV B plasmid[e]	Low positive	60	60	100.0	30.20	0.278	0.9	0.282	0.9	0.396	1.3
	Moderate positive	60	60	100.0	29.39	0.297	1.0	0.548	1.9	0.623	2.1

[a] Total includes Within-Lot and Between-Lot Components.
[b] N is the number of valid replicates.
[c] n is the number of positive replicates, which contribute CN values to the variance component analysis.
[d] HPV 58 was used for the Other HR HPV A panel.
[e] HPV 39 was used for the Other HR HPV B panel.

Table 11. Lot-to-Lot Precision Study: SurePath

Panel	Panel Description	N[b]	n[c]	% Positive	Mean CN	Within-Lot Component SD	Within-Lot Component %CV	Between-Lot Component SD	Between-Lot Component %CV	Total[a] SD	Total[a] %CV
HPV 16 plasmid	Low positive	60	60	100.0	30.50	0.413	1.4	0.437	1.4	0.601	2.0
	Moderate positive	60	60	100.0	29.58	0.294	1.0	0.471	1.6	0.555	1.9
HPV 18 plasmid	Low positive	72	71	98.6	30.06	0.426	1.4	0.000	0.0	0.426	1.4[f]
	Moderate positive	71	71	100.0	29.39	0.355	1.2	0.246	0.8	0.432	1.5
HPV 45 plasmid	Low positive	60	60	100.0	30.32	0.345	1.1	0.000	0.0	0.345	1.1
	Moderate positive	60	60	100.0	29.47	0.281	1.0	0.109	0.4	0.302	1.0
Other HR HPV A plasmid[d]	Low positive	60	59	98.3	30.84	0.351	1.1	0.285	0.9	0.452	1.5[f]
	Moderate positive	60	60	100.0	30.00	0.265	0.9	0.248	0.8	0.363	1.2
Other HR HPV B plasmid[e]	Low positive	60	58	96.7	31.06	0.366	1.2	0.167	0.5	0.402	1.3[f]
	Moderate positive	60	59	98.3	30.44	0.323	1.1	0.161	0.5	0.361	1.2[f]

[a] Total includes Within-Lot and Between-Lot Components
[b] N is the number of valid replicates.
[c] n is the number of positive replicates, which contribute CN values to the variance component analysis.
[d] HPV 58 was used for the Other HR HPV A panel.
[e] HPV 39 was used for the Other HR HPV B panel.
[f] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Reproducibility

Site-to-site reproducibility of the Alinity m HR HPV assay was evaluated with a 22-member panel of samples in ThinPrep PreservCyt Solution and a 13-member panel of samples in SurePath Preservative Fluid. The ThinPrep panel included 6 pools made from clinical specimens collected in ThinPrep PreservCyt Solution and 16 contrived samples prepared with SiHa (HPV 16), HeLa (HPV 18), HPV 45, HPV 39, or HPV 58 plasmids in ThinPrep PreservCyt Solution containing HR HPV negative C33A cells. The SurePath panel included 6 pools made from clinical specimens collected in SurePath Preservative Fluid and 7 contrived samples prepared with SiHa or HeLa in SurePath Preservative Fluid containing HR HPV negative C33A cells. For each sample type, 1 negative clinical pool and 5 moderate positive clinical pools representative of each genotype category (HPV 16, 18, 45, Other HR HPV A, and Other HR HPV B) were prepared. ThinPrep contrived panels consist of high negative, low positive, and moderate positive target levels for each genotype category, in addition to a negative panel member. SurePath contrived panels consist of high negative, low positive, and moderate positive target levels for SiHa and HeLa cell lines, in addition to a negative panel member. Each panel member was tested with 3 lots of Alinity m HR HPV AMP Kit across 3 clinical sites, with one unique lot per site on 5 non-consecutive days. Each site tested 2 runs per day and 4 replicates of each panel member per run on each of the 5 days. Each of the 3 clinical sites used different lots of Alinity m HR HPV CTRL Kits and Alinity m Sample Prep Kit 1. The reproducibility results for ThinPrep panels are summarized in **Table 12** and **Table 13**; the reproducibility results for SurePath panels are summarized in **Table 14** and **Table 15**.

Table 12. Summary of Reproducibility Study Panel Members, Median CN, and Positivity Rates: ThinPrep

Panel	Panel Description	N[a]	Median CN[b]	% Positive (n/N)[c]	% Negative Virus detected (n/N)	% Negative Virus not detected (n/N)
Pooled negative clinical sample	Negative	119	NA	0.0 (0/119)	0.0 (0/119)	100.0 (119/119)
HPV negative cell line	Negative	119	NA	0.0 (0/119)	0.0 (0/119)	100.0 (119/119)
HPV 16 positive SiHa cell line	HPV 16 high negative	118	33.03	50.0 (59/118)	18.6 (22/118)	31.4 (37/118)
HPV 18 positive HeLa cell line	HPV 18 high negative	117	36.00	10.3 (12/117)	1.7 (2/117)	88.0 (103/117)
HPV 45 plasmid	HPV 45 high negative	120	32.17	36.7 (44/120)	59.2 (71/120)	4.2 (5/120)
Other HR HPV A plasmid	Other HR HPV A high negative	119	36.00	18.5 (22/119)	22.7 (27/119)	58.8 (70/119)
Other HR HPV B plasmid	Other HR HPV B high negative	111	32.66	24.3 (27/111)	36.9 (41/111)	38.7 (43/111)
Pooled clinical sample	HPV 16 moderate positive	119	27.72	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)
	HPV 18 moderate positive	119	28.28	98.3 (117/119)	0.0 (0/119)	1.7 (2/119)
	HPV 45 moderate positive	120	28.40	98.3 (118/120)	0.0 (0/120)	1.7 (2/120)
	Other HR HPV A moderate positive	119	28.02	98.3 (117/119)	0.0 (0/119)	1.7 (2/119)
	Other HR HPV B moderate positive	118	26.95	100.0 (118/118)	0.0 (0/118)	0.0 (0/118)
HPV 16 positive SiHa cell line	HPV 16 low positive	118	29.41	100.0 (118/118)	0.0 (0/118)	0.0 (0/118)
	HPV 16 moderate positive	117	28.06	100.0 (117/117)	0.0 (0/117)	0.0 (0/117)
HPV 18 positive HeLa cell line	HPV 18 low positive	120	28.16	96.7 (116/120)	0.8 (1/120)	2.5 (3/120)
	HPV 18 moderate positive	119	27.28	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)
HPV 45 plasmid	HPV 45 low positive	120	30.45	99.2 (119/120)	0.8 (1/120)	0.0 (0/120)
	HPV 45 moderate positive	116	29.89	100.0 (116/116)	0.0 (0/116)	0.0 (0/116)
Other HR HPV A plasmid[d]	Other HR HPV A low positive	119	30.31	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)
	Other HR HPV A moderate positive	118	29.75	100.0 (118/118)	0.0 (0/118)	0.0 (0/118)
Other HR HPV B plasmid[e]	Other HR HPV B low positive	120	30.47	97.5 (117/120)	0.0 (0/120)	2.5 (3/120)
	Other HR HPV B moderate positive	119	28.73	99.2 (118/119)	0.0 (0/119)	0.8 (1/119)

[a] N is the number of valid replicates.

[b] The median CN value reported was calculated for all replicates, regardless whether the CN was ≤ or > the clinical cutoff. Negative replicates with no virus detected were assigned the last cycle of PCR (36.00). Due to 0% virus detection, median CN was not reported for negative panels (NA)

[c] The % Positive was based on the number of valid replicates with a CN value ≤ clinical cutoff.

[d] HPV 58 was used for the Other HR HPV A panel.

[e] HPV 39 was used for the Other HR HPV B panel.

Table 13. Variance Component Analysis Results for Reproducibility Study: ThinPrep

Panel	Panel Description	N[a]	n[b]	Mean CN	Within-Run Component SD	Within-Run Component %CV	Between-Run Component SD	Between-Run Component %CV	Between-Day Component SD	Between-Day Component %CV	Between-Site Component SD	Between-Site Component %CV	Total[c] SD	Total[c] %CV
Pooled clinical sample	HPV 16 moderate positive	119	119	27.47	1.391	5.1	0.000	0.0	0.000	0.0	0.441	1.6	1.459	5.3
	HPV 18 moderate positive	119	117	27.82	1.464	5.3	0.624	2.2	0.184	0.7	0.000	0.0	1.602	5.8[f]
	HPV 45 moderate positive	120	118	28.12	1.355	4.8	0.000	0.0	0.319	1.1	0.189	0.7	1.405	5.0[f]
	Other HR HPV A moderate positive	119	117	27.42	1.506	5.5	0.000	0.0	0.569	2.1	0.115	0.4	1.614	5.9[f]
	Other HR HPV B moderate positive	118	118	26.94	0.550	2.0	0.285	1.1	0.147	0.5	0.035	0.1	0.638	2.4
HPV 16 positive SiHa cell line	HPV 16 low positive	118	118	29.43	0.525	1.8	0.155	0.5	0.177	0.6	0.210	0.7	0.612	2.1
	HPV 16 moderate positive	117	117	28.18	0.525	1.9	0.134	0.5	0.387	1.4	0.148	0.5	0.682	2.4
HPV 18 positive HeLa cell line	HPV 18 low positive	120	116	28.24	0.892	3.2	0.000	0.0	0.000	0.0	0.308	1.1	0.944	3.3[f]
	HPV 18 moderate positive	119	119	27.25	0.667	2.4	0.000	0.0	0.000	0.0	0.000	0.0	0.667	2.4
HPV 45 plasmid	HPV 45 low positive	120	119	30.51	0.435	1.4	0.221	0.7	0.000	0.0	0.054	0.2	0.491	1.6[f]
	HPV 45 moderate positive	116	116	29.90	0.357	1.2	0.065	0.2	0.311	1.0	0.000	0.0	0.478	1.6
Other HR HPV A plasmid[d]	Other HR HPV A low positive	119	119	30.35	0.380	1.3	0.194	0.6	0.124	0.4	0.000	0.0	0.444	1.5
	Other HR HPV A moderate positive	118	118	29.85	0.383	1.3	0.200	0.7	0.324	1.1	0.039	0.1	0.542	1.8
Other HR HPV B plasmid[e]	Other HR HPV B low positive	120	117	30.46	0.391	1.3	0.122	0.4	0.184	0.6	0.102	0.3	0.461	1.5[f]
	Other HR HPV B moderate positive	119	118	28.77	0.376	1.3	0.215	0.7	0.184	0.6	0.149	0.5	0.494	1.7[f]

[a] N is the number of valid replicates.
[b] n is the number of valid positive replicates with a CN value ≤ clinical cutoff.
[c] Total includes Within-Run, Between-Run, Between-Day, and Between-Site Components.
[d] HPV 58 was used for the Other HR HPV A panel.
[e] HPV 39 was used for the Other HR HPV B panel.
[f] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Table 14. Summary of Reproducibility Panel Members, Median CN, and Positivity Rates: SurePath

Panel	Panel Description	N[a]	Median CN[b]	% Positive (n/N)[c]	% Negative Virus detected (n/N)	% Negative Virus not detected (n/N)
Pooled negative clinical sample	Negative	119	NA	0.0 (0/119)	0.0 (0/119)	100.0 (119/119)
HPV negative cell line	Negative	119	NA	0.0 (0/119)	0.0 (0/119)	100.0 (119/119)
HPV 16 positive SiHa cell line	HPV 16 high negative	119	36.00	20.2 (24/119)	0.8 (1/119)	79.0 (94/119)
HPV 18 positive HeLa cell line	HPV 18 high negative	118	36.00	25.4 (30/118)	0.0 (0/118)	74.6 (88/119)
Pooled clinical sample	HPV 16 moderate positive	119	28.52	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)
	HPV 18 moderate positive	117	27.20	99.1 (116/117)	0.9 (1/117)	0.0 (0/117)
	HPV 45 moderate positive	119	28.11	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)
	Other HR HPV A moderate positive	118	27.46	99.2 (117/118)	0.0 (0/118)	0.8 (1/118)
	Other HR HPV B moderate positive	118	28.26	99.2 (117/118)	0.0 (0/118)	0.8 (1/118)
HPV 16 positive SiHa cell line	HPV 16 low positive	120	29.57	100.0 (120/120)	0.0 (0/120)	0.0 (0/120)
	HPV 16 moderate positive	118	27.98	100.0 (118/118)	0.0 (0/118)	0.0 (0/118)
HPV 18 positive HeLa cell line	HPV 18 low positive	118	27.91	98.3 (116/118)	0.0 (0/118)	1.7 (2/118)
	HPV 18 moderate positive	119	26.54	100.0 (119/119)	0.0 (0/119)	0.0 (0/119)

[a] N is the number of valid replicates.
[b] The median CN value reported was calculated for all replicates, regardless whether the CN was ≤ or > the clinical cutoff. Negative replicates with no virus detected were assigned the last cycle of PCR (36.00). Due to 0% virus detection, median CN was not reported for negative panels (NA).
[c] The % Positive was based on the number of valid replicates with a CN value ≤ the clinical cutoff.

Table 15. Variance Component Analysis Results for Reproducibility Study: SurePath

Panel	Panel Description	N[a]	n[b]	Mean CN	Within-Run Component SD	Within-Run Component %CV	Between-Run Component SD	Between-Run Component %CV	Between-Day Component SD	Between-Day Component %CV	Between-Site Component SD	Between-Site Component %CV	Total[c] SD	Total[c] %CV
Pooled clinical sample	HPV 16 moderate positive	119	119	28.44	0.889	3.1	0.000	0.0	0.054	0.2	0.189	0.7	0.911	3.2
	HPV 18 moderate positive	117	116	27.49	1.265	4.6	0.000	0.0	0.256	0.9	0.303	1.1	1.326	4.8[d]
	HPV 45 moderate positive	119	119	28.05	0.785	2.8	0.265	0.9	0.000	0.0	0.181	0.6	0.848	3.0
	Other HR HPV A moderate positive	118	117	27.28	1.110	4.1	0.287	1.1	0.000	0.0	0.045	0.2	1.148	4.2[d]
	Other HR HPV B moderate positive	118	117	28.14	1.102	3.9	0.000	0.0	0.371	1.3	0.542	1.9	1.283	4.6[d]
HPV 16 positive SiHa cell line	HPV 16 low positive	120	120	29.62	0.592	2.0	0.000	0.0	0.077	0.3	0.156	0.5	0.617	2.1
	HPV 16 moderate positive	118	118	28.02	0.439	1.6	0.025	0.1	0.121	0.4	0.051	0.2	0.458	1.6
HPV 18 positive HeLa cell line	HPV 18 low positive	118	116	28.06	0.888	3.2	0.000	0.0	0.059	0.2	0.321	1.1	0.946	3.4[d]
	HPV 18 moderate positive	119	119	26.57	0.636	2.4	0.000	0.0	0.000	0.0	0.066	0.2	0.640	2.4

[a] N is the number of valid replicates.
[b] n is the number of valid positive replicates with a CN value ≤ clinical cutoff.
[c] Total includes Within-Run, Between-Run, Between-Day, and Between-Site Components.
[d] Because only positive results with CN values ≤ the cutoff were included, estimates of SD (and %CV) may be underestimated.

Analytical Specificity

Cross-Reactivity

The cross-reactivity of the Alinity m HR HPV assay was evaluated with a panel of microorganisms that included low-risk HPV, bacteria, viruses, protozoan, and yeast as well as human cellular DNA (**Table 16**). Microorganisms were tested at 10^5 Units/mL for viruses and eukaryotes and 10^6 Units/mL for bacteria, where the unit of measure is specific to each microorganism. Human cellular DNA was tested at 10^6 Copies/mL. Each potential cross reactant was tested with HR HPV negative samples in both ThinPrep PreservCyt Solution and SurePath Preservative Fluid with a minimum of three replicates. No cross-reactivity in Alinity m HR HPV assay performance was observed in the presence of the tested potential cross reactants in either specimen type.

Table 16. Microorganisms Tested

Low Risk HPV			
HPV 6	HPV 34	HPV 54	HPV 70
HPV 11	HPV 40	HPV 55B	HPV 73
HPV 13	HPV 42	HPV 57	HPV 82
HPV 26	HPV 43	HPV 61	HPV 85
HPV 30	HPV 44	HPV 67	
HPV 32	HPV 53	HPV 69	
Bacteria			
Bacteroides fragilis	*Escherichia coli*	*Neisseria gonorrhoeae*	*Streptococcus pneumoniae*
Bacteroides ureolyticus	*Fusobacterium necrophorum*	*Neisseria meningitidis* Serogroup A	*Streptococcus pyogenes*
Bifidobacterium adolescentis	*Gardnerella vaginalis*	*Peptostreptococcus anaerobius*	*Trepenoma pallidum*
Chlamydia trachomatis	*Haemophilus ducreyi*	*Proteus mirabilis*	*Ureaplasma urealyticum*
Clostridium perfringens	*Klebsiella pneumoniae* ss *ozaenae*	*Pseudomonas aeruginosa*	
Corynebacterium genitalium	*Lactobacillus acidophilus*	*Staphylococcus aureus*	
Entercoccus faecalis	*Mycoplasma genitalium*	*Staphylococcus epidermidis*	
Enterobacter cloacae	*Mycoplasma hominis*	*Streptococcus agalactiae*	
Virus			
Adenovirus	Hepatitis B virus (HBV)	Human Herpes virus 3	
Cytomegalovirus (CMV)	Herpes simplex virus I	Hepatitis C virus (HCV)	
Epstein Barr virus (EBV)	Herpes simplex virus II	Human immunodeficiency virus (HIV-1)	
Protozoan	**Yeast**	**Other**	
Trichomonas vaginalis	*Candida albicans*	Human Cellular DNA	

Interference

The effect of potentially interfering endogenous and exogenous substances that may be present in cervical or self-collected vaginal specimens was assessed for the Alinity m HR HPV assay. Substances were tested with HR HPV negative samples (greater than the clinical cutoffs) and HR HPV positive samples containing SiHa (HPV 16) and HeLa (HPV 18) cell lines at approximately 3X LoD in ThinPrep and SurePath clinical cervical matrices and contrived vaginal matrix with a minimum of four replicates.

For both ThinPrep and SurePath, no interference was observed for any substance at the concentrations listed in **Table 17**. Assay interference was observed for Monistat-1 at concentrations higher than 0.75% in ThinPrep. For contrived vaginal matrix, no interference was observed for any substance at the concentrations listed in **Table 18.** Assay interference was observed for Mucus at concentrations higher than 2.5%.

Table 17. Endogenous and Exogenous Substances Evaluated for Cervical Specimens

Endogenous Substance	**Test Level**
Whole Blood	10.00% v/v
Mucus	5.00% w/v[a]
PBMC (leukocytes)	10^6 Cells/mL
Exogenous Substance	**Test Level**
Clotrimazole Vaginal Cream	1.00% w/v
Terconazole Vaginal Cream 0.8%	1.00% w/v
Monistat-1	0.75% w/v[b]
KY Jelly Personal Lubricant	1.00% w/v
KY Warming Liquid	1.00% w/v
Zovirax (Acyclovir)	1.00% w/v
Summer's Eve Douche	1.00% w/v
Norforms Deodorant Suppositories	0.50% w/v
Metrogel-Vaginal	0.50% w/v
Vaginal Contraceptive Foam (VCF) Contraceptive Gel	0.50% w/v
Options Gyncol II Vaginal Contraceptive Gel	0.50% w/v
Hydrocortisone	0.50% w/v
Sigma Acetic Acid	0.50% w/v
Vagisil Dry Wash	0.50% w/v

[a] Based on reported CN values, detection of HPV may be impacted for cervical specimens stored in SurePath in the presence of mucus at a concentration of 5.00% w/v.

[b] 1.00% w/v produced false negative results in ThinPrep clinical matrix.

Table 18. Endogenous and Exogenous Substances Evaluated for Vaginal Specimens

Endogenous Substance	**Test Level**
Whole Blood	10.00% v/v
Mucus	2.50% w/v[a]
PBMC (leukocytes)	10^6 Cells/mL
Semen	5.00% v/v
Estradiol	1.30 ng/mL
Progesterone	20 ng/mL
Exogenous Substance	**Test Level**
Clotrimazole Vaginal Cream	1.00% w/v
Terconazole-3 Vaginal Cream 0.8%	1.00% w/v
Monistat-1	1.00% w/v
KY Jelly Personal Lubricant	1.00% w/v
KY Warming Liquid	1.00% w/v
Zovirax (Acyclovir)	1.00% w/v
Summer's Eve Medicated Douche	1.00% w/v
Norforms Deodorant Suppositories	0.50% w/v
Metronidazole Gel	0.50% w/v
Vaginal Contraceptive Foam (VCF) Concentrative Gel	0.50% w/v
Hydrocortisone	0.50% w/v
Preparation-H	0.50% w/v
Vagisil Dry Wash	0.50% w/v

[a] 5.00% w/v caused assay interference

Competitive Inhibition

Competitive inhibition was assessed with ThinPrep and SurePath samples containing HPV 16, HPV 18, HPV 45, HPV 39, or HPV 58 at low concentration in the presence of 10^5 Copies/mL of each of the other HR HPV genotypes (HPV 16, HPV 18, HPV 45, HPV 39, or HPV 58).

For ThinPrep, no competitive inhibition was observed for any tested genotype at low concentration. For SurePath, no competitive inhibition was observed for HPV 16, HPV 18, or HPV 45 at low concentration. For HPV 58 (Other HR HPV A), no competitive inhibition was observed in the presence of 10^5 Copies/mL of HPV 16, HPV 45, or HPV 39 (Other HR HPV B). Detection of HPV 58 was inhibited in the presence of 10^5 Copies/mL of HPV 18 but not 10^4 Copies/mL. For HPV 39 (Other HR HPV B), no competitive inhibition was observed in the presence of high concentrations of HPV 58 (Other HR HPV A). Detection of HPV 39 (Other HR HPV B) was inhibited in the presence of 10^5 Copies/mL of HPV 16, HPV 18, or HPV 45 but not 10^4 Copies/mL.

Carryover

Carryover studies were conducted to stress two areas of potential carryover on the Alinity m System: Sample Preparation and the AMP TRAY 1. Carryover rates were determined for each by testing alternating replicates of HR HPV high positive samples and HR HPV negative samples across multiple runs. The HR HPV high positive sample was prepared with HPV 16 at 6.9×10^7 Copies/mL.

While no carryover occurred in the second study (AMP TRAY 1), the highest potential for carryover was determined to be at the Sample Preparation stage. Of 481 negative samples tested adjacent to high positive samples during sample preparation, 4 samples were positive in the Sample Preparation study, resulting in an overall carryover rate of 0.83% (4/481, 95%CI: 0.32% to 2.12%).

CLINICAL PERFORMANCE EVALUATION

Expected Results

A multi-center, prospective study was conducted to evaluate the performance of Alinity m HR HPV. The study recruited 11,711 women ≥ 25 years of age who were undergoing routine cervical cancer screening (Screening population) in the US across 96 sites. Included in the study were 11,532 women with valid Alinity m HR HPV assay results in ThinPrep (11,521 results) and SurePath (9,466 results). The Alinity m HR HPV assay testing was performed at 4 clinical testing sites.

In the Screening population, the median age of the women was 41, with 13.4% in age group 25-29 years, 30.9% in age group 30-39 years, and 55.7% in age group ≥ 40 years. The percent of invalid Alinity m HR HPV test results in ThinPrep was 0.06% (7/11,528) with 95% CI: 0.03% to 0.13% and in SurePath was 1.14% (109/9,575) with a 95% CI: 0.94% to 1.37%.

The Alinity m HR HPV assay valid results for genotype/genotype groups stratified by specimen type and age group is outlined in **Table 19** for the Screening population.

Table 19. The Alinity m HR HPV Assay Genotype/Genotype Group Result by Specimen Type and Age Group for the Screening Population

Specimen Type	Age Group	HPV 16+ (n/N)	HPV 18+ (n/N)	HPV 45+ (n/N)	Other HR HPV A+ (n/N)	Other HR HPV B+ (n/N)	Negative (n/N)
ThinPrep	25-29	1.2% (18/1546)	0.3% (5/1546)	1.4% (21/1546)	5.8% (89/1546)	11.7% (181/1546)	81.8% (1265/1546)
	30-39	1.3% (46/3558)	0.9% (31/3558)	1.2% (42/3558)	4.6% (163/3558)	6.3% (225/3558)	87.2% (3104/3558)
	≥40	1.1% (68/6417)	0.6% (40/6417)	0.7% (45/6417)	2.6% (167/6417)	4.1% (263/6417)	91.7% (5885/6417)
	Total	1.1% (132/11521)	0.7% (76/11521)	0.9% (108/11521)	3.6% (419/11521)	5.8% (669/11521)	89.0% (10254/11521)
SurePath	25-29	1.3% (17/1303)	0.2% (3/1303)	1.5% (20/1303)	5.4% (70/1303)	11.6% (151/1303)	81.9% (1067/1303)
	30-39	1.2% (36/2907)	1.0% (30/2907)	1.1% (33/2907)	4.7% (137/2907)	6.1% (178/2907)	87.2% (2534/2907)
	≥40	1.2% (61/5256)	0.6% (31/5256)	0.8% (43/5256)	2.9% (152/5256)	4.7% (247/5256)	90.8% (4775/5256)
	Total	1.2% (114/9466)	0.7% (64/9466)	1.0% (96/9466)	3.8% (359/9466)	6.1% (576/9466)	88.5% (8376/9466)

Table 20 shows HPV positivity rate among valid results for the Alinity m HR HPV assay by specimen type and age group. HPV positivity rate decreases with age when tested with either ThinPrep or SurePath.

Table 20. HPV Positivity Rate as Determined by the Alinity m HR HPV Assay by Specimen Type and Age Group in the Screening Population

Specimen Type	Age Group	Positivity Rate (n/N)
ThinPrep	25-29	18.2% (281/1546)
	30-39	12.8% (454/3558)
	≥40	8.3% (532/6417)
	Total	11.0% (1267/11521)
SurePath	25-29	18.1% (236/1303)
	30-39	12.8% (373/2907)
	≥40	9.2% (481/5256)
	Total	11.5% (1090/9466)

Table 21 shows HPV positivity rate among valid results for the Alinity m HR HPV assay by specimen type and testing site. The overall HPV positivity rate was 11.0% and 11.5% for ThinPrep and SurePath specimens, respectively.

Table 21. HPV Positivity Rate as Determined by the Alinity m HR HPV Assay by Specimen Type and Testing Site in the Screening Population

Specimen Type	Testing Site	Positivity Rate (n/N)
ThinPrep	1	10.6% (260/2442)
	2	10.8% (501/4643)
	3	11.7% (280/2397)
	4	11.1% (226/2039)
	Total	11.0% (1267/11521)
SurePath	1	11.4% (187/1639)
	2	11.1% (382/3449)
	3	11.6% (263/2261)
	4	12.2% (258/2117)
	Total	11.5% (1090/9466)

Clinical Study Design

The study recruited 14,935 women ≥ 25 years in the US across 96 collection sites, including 11,711 undergoing routine cervical cancer screening (Screening population) and 3,224 referred for colposcopy follow-up based on previous cervical cancer screening that included HR HPV testing (Enriched population). Screening and Enriched populations include women in the US from across 96 collection sites. The Alinity m HR HPV assay testing was performed at 4 clinical testing sites. All cytology and HPV testing by another FDA-approved HPV test were performed at a reference laboratory.

Screening Population

The collection sites served populations at low to high risks for HPV infection and included but were not limited to health clinics, OB/GYN practices, health integrated site networks/health systems, private practice and research centers, and universities and academic centers. A total of 11,636 women were eligible to participate in the study.

All women in the Screening population had cervical specimens collected using an endocervical brush/spatula and placed in ThinPrep PreservCyt Solution (ThinPrep). Approximately 80% of the women also had cervical specimens collected using a cervical broom-like collection device and placed in SurePath Preservative Fluid (SurePath). When both cervical specimens were collected, the collection order were randomized to ensure that approximately half of the women had ThinPrep collected first, and the other half had SurePath collected first.

In the Screening population, specimens collected as the first (or only) specimen were used for cytology testing also. ThinPrep specimens were processed for cytology using the ThinPrep 2000 (TP 2000) or ThinPrep 5000 (TP 5000), and SurePath specimens were processed using the BD PrepMate/PrepStain. ThinPrep specimens were randomized to ensure that approximately half were processed with TP 2000, and the remaining half with TP 5000. The slides generated from TP 2000, TP 5000 and PrepMate/PrepStain Systems were diagnosed using the 2014 Bethesda System.[37] All specimens collected were tested for HPV by Alinity m HR HPV and an FDA-approved HPV test performed according to manufacturer's instructions. For ThinPrep, HPV testing (Alinity m HR HPV and FDA-approved tests) was performed using an aliquot taken prior to cytology processing (pre-quot) or without subsequent cytology processing (non-cytology aliquot) for ThinPrep specimens collected as a second sample. For SurePath, when SurePath was collected first, the Alinity m HR HPV assay testing was performed on pre-quot specimens while the FDA-approved HPV test was performed after cytology processing (post-quot). When SurePath was collected second, both the Alinity m HR HPV and the FDA-approved HPV tests were performed on non-cytology aliquots. Cytology results and up to four HPV results (from both the Alinity m HR HPV assay and the FDA-approved HPV test and both ThinPrep and SurePath specimens) were utilized to inform referral to colposcopy. Women in the Screening population with ≥ ASC-US cytology regardless of HPV result or with any cytology result and a positive HPV status (any positive from up to 4 results) were referred to colposcopy. Those with a NILM cytology and negative or undetermined HPV, or negative HPV and UNSAT or unavailable cytology, were not referred to colposcopy. For women not referred to colposcopy, when cytology was UNSAT or not done and HPV status was undetermined (due to missing HPV result(s)), the disease status was considered undetermined. Women with NILM (normal) cytology and/or negative HPV status (all HPV results were negative) were also considered disease negative. Summary of Alinity m HR HPV results by cytology for the Screening population is shown in **Table 22** and **Table 23** for ThinPrep and SurePath, respectively.

Table 22. The Alinity m HR HPV Assay Results by Cytology Result for Screening Population – ThinPrep

	Cytology Result				
Alinity m HR HPV[a]	**> ASC-US**	**ASC-US**	**Normal[b]**	**UNSAT/Unknown**	**Total**
HPV 16	22	11	96	3	132
HPV 18	11	14	49	2	76
HPV 45	11	11	81	5	108
Other HR HPV A	48	56	305	10	419
Other HR HPV B	99	78	480	12	669
Negative	95	388	9500	271	10254

[a] Detection of more than 1 HR HPV genotype/genotype group is counted separately.

[b] Subjects with cytology result as NILM and "Other: Endometrial cells (in women aged ≥ 45 years)" are included in Normal Category.

Table 23. The Alinity m HR HPV Assay Results by Cytology Result for Screening Population – SurePath

	Cytology Result				
Alinity m HR HPV[a]	**> ASC-US**	**ASC-US**	**Normal[b]**	**UNSAT/Unknown**	**Total**
HPV 16	21	8	84	1	114
HPV 18	7	12	45	0	64
HPV 45	9	10	73	4	96
Other HR HPV A	37	46	269	7	359
Other HR HPV B	79	68	423	6	576
Negative	82	306	7831	157	8376

[a] Detection of more than 1 HR HPV genotype/genotype group is counted separately.

[b] Subjects with cytology result as NILM and "Other: Endometrial cells (in women aged ≥ 45 years)" are included in Normal Category.

Enriched Population

All women in the Enriched population had both ThinPrep and SurePath specimens collected. Women referred for colposcopy follow-up based on previous cervical cancer screening that included HR HPV testing represented the Enriched population. For women in the Enriched population, specimens collected were tested by both the Alinity m HR HPV assay and the FDA-approved HPV test using non-cytology aliquots. All women in the Enriched population underwent colposcopy procedures.

Colposcopy and Histology Process

Colposcopy procedures were conducted in Screening and Enriched populations according to a standardized protocol: 1) an endocervical curettage was performed for all participants; 2) biopsies were obtained on all visible lesions, and 3) a single random biopsy of the visualized or partially visualized squamocolumnar junction was obtained if no lesion was visible. In addition, during the colposcopy visit, LEEP was performed when required by the standard of care. To avoid potential bias, study participants and colposcopists were blinded to HPV test results until colposcopy was completed. Cytology results were made available to colposcopists prior to colposcopy.

All tissue biopsies were examined by a Central Pathology Review Panel (CPRP) consisting of 3 expert pathologists. Discordant results were adjudicated according to a pre-defined standardized protocol. The CPRP was masked to patient information, including cytology, HPV results, and previous diagnoses. The slides prepared from the tissue biopsies were stained using conventional hematoxylin and eosin (H&E) staining, and with p16 IHC staining where applicable. For all tissue biopsies, the expert pathologists evaluated H&E slides to establish an H&E histologic diagnosis. When a tissue biopsy case met LAST (Lower Anogenital Squamous Terminology) Workshop guidelines, the expert pathologists evaluated both p16 and H&E slides for that tissue biopsy to establish a p16/H&E histologic diagnosis. The final CPRP histologic results for the biopsies meeting the LAST guidelines

were defined by the p16/H&E histologic diagnosis. The final CPRP histologic results for the biopsies not meeting the LAST guidelines were defined by the H&E histologic diagnosis. After the slides from all collected tissues were reviewed for a woman, the disease status for the woman was determined based on the most severe histologic result across all tissues collected (≥ CIN3, CIN2, or ≤ CIN1).

Alinity m HR HPV Assay Performance for ≥ CIN3 and ≥ CIN2 Clinical Endpoints

The clinical performance was evaluated for the Screening and Enriched populations separately for ≥ CIN3 and ≥ CIN2 clinical endpoints. For the ≥ CIN3 endpoint, the disease positive women included those with either CIN3 or cancer (including invasive cervical cancer, carcinoma in situ, and adenocarcinoma in situ); the disease negative women (< CIN3) included those with histologic result categories of either negative, CIN1, or CIN2. For the ≥ CIN2 endpoint, the disease positive women included those with either CIN2, CIN3, or cancer; the disease negative (< CIN2) women included those with histologic result categories of either negative or CIN1.

A positive test result is defined as HR HPV Positive by the Alinity m HR HPV assay (reporting either HPV 16, 18, HPV 45, Group A, or Group B) and/or FDA-approved HPV test (reported as HPV 16, 18, or 12 Other). The positive test results summarized below in the Screening and Enriched populations are reported for both the Alinity m HR HPV and FDA-approved tests as HPV 16/18 combined and 12 Other, encompassing the remaining HR HPV genotypes.

Screening Population

The performance of the Alinity m HR HPV assay was assessed based on the ratios of clinical sensitivities and the ratios of false positive rates between Alinity m HR HPV and the FDA-approved HPV test for both ≥ CIN3 and ≥ CIN2 clinical endpoints. Ratios were assessed as only women with either positive HPV status or cytology result ≥ ASC-US were referred to colposcopy. The two-sided 95% confidence intervals for the ratios of clinical sensitivities and the ratios of false positive rates were calculated.

Enriched Population

Because all women in the Enriched population have histology results, clinical sensitivity and false positive rate of Alinity m HR HPV and the FDA-approved tests were calculated along with two-sided 95% CI. In addition, the ratios of clinical sensitivities and the ratios of false positive rates between Alinity m HR HPV and the FDA-approved tests were calculated along with two-sided 95% CIs.

The cross-tabulation of the Alinity m HR HPV and FDA-approved tests against ≥ CIN2 and ≥ CIN3 disease endpoints, in both ThinPrep and SurePath matrices, are present below for the Screening (**Table 24** through **Table 27**) and Enriched (**Table 29** through **Table 32**) populations. The final estimated clinical sensitivity and false positive rate, in addition to the ratio comparing Alinity m HR HPV and the FDA-approved tests are presented for ThinPrep and SurePath in both Screening (**Table 28**) and Enriched (**Table 33**) populations below.

Clinical Performance Characteristics in the Screening Population

Table 24. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN2 endpoint: ThinPrep Screening population

	≥CIN2				<CIN2			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	**HPV 16/18**	**12 Other**	**Negative**	**Total**	**HPV 16/18**	**12 Other**	**Negative**	**Total**
HPV 16/18	29	0	0	29	102	1	15	118
12 Other	1	68	6	75	11	569	104	684
Negative	1	2	7	10	11	87	9942	10040
Total	31	70	13	114	124	657	10061	10842

Table 25. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN3 endpoint: ThinPrep Screening population

	≥CIN3				<CIN3			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	**HPV 16/18**	**12 Other**	**Negative**	**Total**	**HPV 16/18**	**12 Other**	**Negative**	**Total**
HPV 16/18	13	0	0	13	118	1	15	134
12 Other	0	20	0	20	12	617	110	739
Negative	0	0	2	2	12	89	9947	10048
Total	13	20	2	35	142	707	10072	10921

Table 26. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN2 endpoint: SurePath Screening population

	≥CIN2				<CIN2			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	**HPV 16/18**	**12 Other**	**Negative**	**Total**	**HPV 16/18**	**12 Other**	**Negative**	**Total**
HPV 16/18	17	0	1	18	86	3	15	104
12 Other	2	54	3	59	6	482	73	561
Negative	1	3	9	13	8	114	7578	7700
Total	20	57	13	90	100	599	7666	8365

Table 27. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN3 endpoint: SurePath Screening population

	≥CIN3				<CIN3			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	**HPV 16/18**	**12 Other**	**Negative**	**Total**	**HPV 16/18**	**12 Other**	**Negative**	**Total**
HPV 16/18	8	0	0	8	95	3	16	114
12 Other	1	16	0	17	7	520	76	603
Negative	0	0	2	2	9	117	7585	7711
Total	9	16	2	27	111	640	7677	8428

Table 28. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN2 and ≥CIN3 endpoints: Screening population

		Clinical sensitivity				False positive rate		
Specimen Type	Clinical endpoint	Alinity m HR HPV (95% CI)	FDA-approved Comparator (95% CI)	Ratio (95% CI)	Clinical endpoint	Alinity m HR HPV (95% CI)	FDA-approved Comparator (95% CI)	Ratio (95% CI)
ThinPrep	≥CIN2	91.2% (104/114) (84.6%, 95.2%)	88.6% (101/114) (81.5%, 93.2%)	1.03 (0.97, 1.09)	<CIN2	7.4% (802/10842) (6.9%, 7.9%)	7.2% (781/10842) (6.7%, 7.7%)	1.03 (0.99, 1.07)
	≥CIN3	94.3% (33/35) (81.4%, 98.4%)	94.3% (33/35) (81.4%, 98.4%)	1.00 NA	<CIN3	8.0% (873/10921) (7.5%, 8.5%)	7.8% (849/10921) (7.3%, 8.3%)	1.03 (0.99, 1.06)
SurePath	≥CIN2	85.6% (77/90) (76.8%; 91.4%)	85.6% (77/90) (76.8%; 91.4%)	1.00 (0.93, 1.07)	<CIN2	7.9% (665/8365) (7.4%, 8.5%)	8.4% (699/8365) (7.8%, 9.0%)	0.95 (0.91, 0.99)
	≥CIN3	92.6% (25/27) (76.6%, 97.9%)	92.6% (25/27) (76.6%, 97.9%)	1.00 NA	<CIN3	8.5% (717/8428) (7.9%, 9.1%)	8.9% (751/8428) (8.3%, 9.5%)	0.95 (0.91, 0.99)

NA: The 95% CI was not calculated as Alinity m HR HPV and FDA-approved tests have the same results for every specimen.

Clinical Performance Characteristics in the Enriched Population

Table 29. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN2 endpoint: ThinPrep Enriched population

	≥CIN2				<CIN2			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	HPV 16/18	12 Other	Negative	Total	HPV 16/18	12 Other	Negative	Total
HPV 16/18	189	0	2	191	233	4	14	251
12 Other	5	347	4	356	18	1251	122	1391
Negative	0	9	28	37	5	99	651	755
Total	194	356	34	584	256	1354	787	2397

Table 30. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN3 endpoint: ThinPrep Enriched population

	≥CIN3				<CIN3			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	HPV 16/18	12 Other	Negative	Total	HPV 16/18	12 Other	Negative	Total
HPV 16/18	87	0	1	88	335	4	15	354
12 Other	0	86	1	87	23	1510	125	1658
Negative	0	1	8	9	5	107	671	783
Total	87	87	10	184	363	1621	811	2795

Table 31. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN2 endpoint: SurePath Enriched population

	≥CIN2				<CIN2			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	HPV 16/18	12 Other	Negative	Total	HPV 16/18	12 Other	Negative	Total
HPV 16/18	190	0	0	190	247	2	10	259
12 Other	9	338	2	349	23	1248	94	1365
Negative	0	18	25	43	6	126	593	725
Total	199	356	27	582	276	1376	697	2349

Table 32. Cross-tabulation of Alinity m HR HPV and FDA-approved test results at the ≥CIN3 endpoint: SurePath Enriched population

	≥CIN3				<CIN3			
	FDA-approved test				FDA-approved test			
Alinity m HR HPV	HPV 16/18	12 Other	Negative	Total	HPV 16/18	12 Other	Negative	Total
HPV 16/18	87	0	0	87	350	2	10	362
12 Other	0	84	0	84	32	1500	96	1628
Negative	0	4	8	12	6	140	610	756
Total	87	88	8	183	388	1642	716	2746

Table 33. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN2 and ≥CIN3 endpoints: Enriched population

		Clinical sensitivity				False positive rate[b]		
Specimen Type	Clinical endpoint	Alinity m HR HPV (95% CI)	FDA-approved Comparator (95% CI)	Ratio[a] (95% CI)	Clinical endpoint	Alinity m HR HPV (95% CI)	FDA-approved Comparator (95% CI)	Ratio (95% CI)
ThinPrep	≥CIN2	93.7% (547/584) (91.4%, 95.4%)	94.2% (550/584) (92.0%, 95.8%)	0.99 (0.98, 1.01)	<CIN2	68.5% (1642/2397) (66.6%, 70.3%)	67.2% (1610/2397) (65.3%, 69.0%)	1.02 (1.00, 1.04)
	≥CIN3	95.1% (175/184) (91.0%, 97.4%)	94.6% (174/184) (90.3%, 97.0%)	1.01 (0.99, 1.02)	<CIN3	72.0% (2012/2795) (70.3%, 73.6%)	71.0% (1984/2795) (69.3%, 72.6%)	1.01 (1.00, 1.03)
SurePath	≥CIN2	92.6% (539/582) (90.2%, 94.5%)	95.4% (555/582) (93.3%, 96.8%)	0.97 (0.96, 0.99)	<CIN2	69.1% (1624/2349) (67.2%, 71.0%)	70.3% (1652/2349) (68.4%, 72.1%)	0.98 (0.97, 1.00)
	≥CIN3	93.4% (171/183) (88.9%, 96.2%)	95.6% (175/183) (91.6%, 97.8%)	0.98 (0.95, 0.99)	<CIN3	72.5% (1990/2746) (70.8%, 74.1%)	73.9% (2030/2746) (72.3%, 75.5%)	0.98 (0.96, 1.00)

[a] The ratio reported falls within performance range of FDA-approved HPV devices compared with one another.
[b] The false positive rate reported for both the Alinity m HR HPV and FDA-approved test was high in the Enriched population as this was a biased colposcopy referral population with likely high viral loads related to disease. However, false positive rates of Alinity m HR HPV and the FDA-approved tests were similar.

The overall ratio of clinical sensitivities between Alinity m HR HPV and the FDA-approved HPV test for detecting disease (≥CIN3) was 1.00 for ThinPrep and 0.98 for SurePath. For ThinPrep, the ratio of clinical sensitivities was 1.00 for the Screening population and 1.01 for the Enriched population. For SurePath, the ratio of clinical sensitivities was 1.00 for the Screening population and 0.98 for the Enriched population. The overall ratio of clinical sensitivities between Alinity m HR HPV and the FDA-approved HPV test for detecting disease (≥CIN2) was 1.00 for ThinPrep and 0.97 for SurePath. For ThinPrep, the ratio of clinical sensitivities was 1.03 for the Screening population and 0.99 for the Enriched population. For SurePath, the ratio of clinical sensitivities was 1.00 for the Screening population and 0.97 for the Enriched population.

Agreement with HPV DNA Sequencing Results

The Alinity m HR HPV genotype specific agreement was evaluated by comparing the Alinity m HR HPV assay results with HPV DNA next-generation sequencing (NGS).

A representative subset of the ThinPrep and SurePath pre-quot or non-cytology aliquots from the Screening population were tested with HPV DNA NGS at a reference laboratory. The agreement (PPA, NPA, and 95% CI) between Alinity m HR HPV and NGS genotyping results were estimated for HPV virus detected (CN ≤ or > clinical cutoff) and positive (CN ≤ clinical cutoff) specimens for each respective channel. The individual Alinity m HR HPV genotype results are summarized for each specimen type (ThinPrep and SurePath) and are presented in **Table 34** through **Table 38** for ThinPrep and **Table 39** through **Table 43** for SurePath in the Screening population.

Screening Population: ThinPrep Agreements

Table 34. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 16 Detection: ThinPrep Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	23	12	0	35				
Negative, virus detected	0	1	0	1	95.83	98.22	95.83	98.35
Negative, virus not detected	1	716	5	722	(23/24)	(716/729)	(23/24)	(717/729)
Total	24	729	5	758	(79.76, 99.26)	(96.97, 98.95)	(79.76, 99.26)	(97.14, 99.06)

Table 35. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 18 Detection: ThinPrep Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	14	7	0	21				
Negative, virus detected	0	0	0	0	100.00	99.05	100.00	99.05
Negative, virus not detected	0	732	5	737	(14/14)	(732/739)	(14/14)	(732/739)
Total	14	739	5	758	(78.47, 100.00)	(98.06, 99.54)	(78.47, 100.00)	(98.06, 99.54)

Table 36. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 45 Detection: ThinPrep Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	24	10	0	34				
Negative, virus detected	0	1	0	1	92.31%	98.49	92.31	98.62
Negative, virus not detected	2[a]	716	5	723	(24/26)	(716/727)	(24/26)	(717/727)
Total	26	727	5	758	(75.86, 97.86)	(97.31, 99.15)	(75.86, 97.86)	(97.49, 99.25)

[a] Of the 2 cases, the histology results included 1 negative and 1 undetermined due to inadequate histology specimen with no visible lesion by colposcopic review.

Table 37. Agreement Between Alinity m HR HPV and HPV DNA NGS for Other HR HPV A Detection: ThinPrep Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	88	22	0	110				
Negative, virus detected	2	0	0	2	87.38	96.62	85.44	96.62
Negative, virus not detected	13[a]	628	5	646	(90/103)	(628/650)	(88/103)	(628/650)
Total	103	650	5	758	(79.60, 92.47)	(94.93, 97.75)	(77.35, 90.97)	(94.93, 97.75)

[a] Of the 13 cases, the histology results included 1 > CIN3, 1 CIN2, 2 CIN1, 8 negative and 1 undetermined due to loss to follow up. The > CIN3 case had NGS result of HPV 45/52, Alinity m HR HPV result of HPV 45, and comparator result of Other HR HPV. The CIN2 case had NGS result of HPV52/35/56, Alinity m HR HPV result of Other HR HPV B and comparator result of Other HR HPV.

Table 38. Agreement Between Alinity m HR HPV and HPV DNA NGS for Other HR HPV B Detection: ThinPrep Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	150	27	0	177				
Negative, virus detected	7	3	0	10	91.28	94.84	87.21	95.35
Negative, virus not detected	15[a]	551	5	571	(157/172)	(551/581)	(150/172)	(554/581)
Total	172	581	5	758	(86.11, 94.64)	(92.72, 96.36)	(81.39, 91.40)	(93.32, 96.79)

[a] Of the 15 cases, the histology results included 1 CIN2, 1 CIN1, 7 negative, and 6 undetermined due to loss to follow up. The CIN2 case had NGS result of HPV 51 and negative comparator result.

Screening Population: SurePath Agreements

Table 39. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 16 Detection: SurePath Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	22	17	0	39				
Negative, virus detected	0	1	0	1	95.65 (22/23) (79.01, 99.23)	97.49 (698/716) (96.06, 98.40)	95.65 (22/23) (79.01, 99.23)	97.63 (699/716) (96.23, 98.51)
Negative, virus not detected	1	698	5	704				
Total	23	716	5	744				

Table 40. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 18 Detection: SurePath Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	14	7	0	21				
Negative, virus detected	0	0	0	0	100.00 (14/14) (78.47, 100.00)	99.03 (718/725) (98.02, 99.53)	100.00 (14/14) (78.47, 100.00)	99.03 (718/725) (98.02, 99.53)
Negative, virus not detected	0	718	5	723				
Total	14	725	5	744				

Table 41. Agreement Between Alinity m HR HPV and HPV DNA NGS for HPV 45 Detection: SurePath Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	21	13	0	34				
Negative, virus detected	1	1	0	2	88.00 (22/25) (70.04, 95.83)	98.04 (700/714) (96.74, 98.83)	84.00 (21/25) (65.35, 93.60)	98.18 (701/714) (96.91, 98.93)
Negative, virus not detected	3[a]	700	5	708				
Total	25	714	5	744				

[a] Of the 3 cases, the histology results included 1 negative, 2 undetermined due to loss to follow up or inadequate histology specimen with no visible lesion by colposcopic review.

Table 42. Agreement Between Alinity m HR HPV and HPV DNA NGS for Other HR HPV A Detection: SurePath Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	80	32	0	112				
Negative, virus detected	1	1	0	2	90.00 (81/90) (82.08, 94.65)	94.92 (616/649) (92.95, 96.36)	88.89 (80/90) (80.74, 93.85)	95.07 (617/649) (93.12, 96.49)
Negative, virus not detected	9[a]	616	5	630				
Total	90	649	5	744				

[a] Of the 9 cases, the histology results included 1 > CIN3, 2 CIN2, 2 CIN1, 3 negative and 1 undetermined due to loss to follow up. The > CIN3 case had NGS result of HPV 45/52, Alinity m HR HPV result of HPV 45 and comparator result of Other HR HPV. The 2 CIN2 cases had NGS result of HPV 52/59 and 52/35/56, Alinity m HR HPV result of Other HR HPV B and comparator result of Other HR HPV.

Table 43. Agreement Between Alinity m HR HPV and HPV DNA NGS for Other HR HPV B Detection: SurePath Screening

	NGS Result				Detected (CN ≤ or > Clinical Cutoff)		Positive (CN ≤ Clinical Cutoff)	
Alinity m HR HPV	Positive	Negative	Invalid	Total	PPA (%) (95% CI)	NPA (%) (95% CI)	PPA (%) (95% CI)	NPA (%) (95% CI)
Positive	137	36	0	173				
Negative, virus detected	4	11	0	15	90.97 (141/155) (85.41, 94.54)	91.95 (537/584) (89.46, 93.89)	88.39 (137/155) (82.39, 92.53)	93.84 (548/584) (91.58, 95.51)
Negative, virus not detected	14[a]	537	5	556				
Total	155	584	5	744				

[a] Of the 14 cases, the histology results included 1 CIN2, 11 negative and 2 undetermined due to non-referral to colposcopy or loss to follow up. The CIN2 case had NGS result of HPV 51 and negative comparator result.

In the Screening population, the Alinity m HR HPV genotype agreements ranged from 85.44% to 100% in ThinPrep and 84.00% to 100% in SurePath when evaluated using the clinical cutoff and ranged from 87.38% to 100% in ThinPrep and 88.00% to 100% in SurePath when evaluated for HPV virus detection (CN ≤ or > the clinical cutoff).

Performance by HPV Vaccination Status

In the Screening and Enriched populations, the performance of the Alinity m HR HPV assay for vaccinated and unvaccinated subjects was assessed based on the ratios of clinical sensitivities and the ratios of false positive rates between Alinity m HR HPV and the FDA-approved HPV test, for both ≥ CIN3 and ≥ CIN2 clinical endpoints. The two-sided 95% confidence intervals for the ratios of clinical sensitivities and the ratios of false positive rates were calculated. Refer to **Table 44** through **Table 47**.

Table 44. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN2 endpoints by vaccination status: Screening population

		Clinical sensitivity			False positive rate		
Specimen Type	**Status**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**
ThinPrep	Vaccinated	95.5% (21/22) (78.2%, 99.2%)	90.9% (20/22) (72.2%, 97.5%)	1.05 (0.90, 1.24)	10.0% (144/1441) (8.5%, 11.6%)	10.3% (148/1441) (8.8%, 11.9%)	0.97 (0.90, 1.04)
	Unvaccinated	88.8% (71/80) (80.0%, 94.0%)	87.5% (70/80) (78.5%, 93.1%)	1.01 (0.96, 1.09)	6.6% (570/8616) (6.1%, 7.2%)	6.5% (558/8616) (6.0%, 7.0%)	1.02 (0.98, 1.07)
SurePath	Vaccinated	85.0% (17/20) (64.0%, 94.8%)	80.0% (16/20) (58.4%, 91.9%)	1.06 (0.82, 1.43)	10.4% (120/1159) (8.7%, 12.2%)	11.3% (131/1159) (9.6%, 13.3%)	0.92 (0.84, 1.00)
	Unvaccinated	83.1% (49/59) (71.5%, 90.5%)	84.7% (50/59) (73.5%, 91.8%)	0.98 (0.90, 1.04)	7.1% (466/6573) (6.5%, 7.7%)	7.4% (485/6573) (6.8%, 8.0%)	0.96 (0.91, 1.01)

Table 45. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN3 endpoints by vaccination status: Screening population

		Clinical sensitivity			False positive rate		
Specimen Type	**Status**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**
ThinPrep	Vaccinated	100.0% (5/5) (56.6%, 100.0%)	100.0% (5/5) (56.6%, 100.0%)	1.00 NA	11.0% (160/1458) (9.5%, 12.7%)	11.2% (163/1458) (9.7%, 12.9%)	0.98 (0.92, 1.05)
	Unvaccinated	92.9% (26/28) (77.4%, 98.0%)	92.9% (26/28) (77.4%, 98.0%)	1.00 NA	7.1% (615/8668) (6.6%, 7.7%)	6.9% (602/8668) (6.4%, 7.5%)	1.02 (0.98, 1.07)
SurePath	Vaccinated	100.0% (5/5) (56.6%, 100.0%)	100.0% (5/5) (56.6%, 100.0%)	1.00 NA	11.2% (132/1174) (9.6%, 13.2%)	12.1% (142/1174) (10.4%, 14.1%)	0.93 (0.85, 1.01)
	Unvaccinated	90.5% (19/21) (71.1%, 97.3%)	90.5% (19/21) (71.1%, 97.3%)	1.00 NA	7.5% (496/6611) (6.9%, 8.2%)	7.8% (516/6611) (7.2%, 8.5%)	0.96 (0.91, 1.01)

NA: The 95% CI was not calculated as Alinity m HR HPV and FDA-approved test have the same results for every specimen.

Table 46. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN2 endpoints by vaccination status: Enriched population

		Clinical sensitivity			False positive rate		
Specimen Type	**Status**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**
ThinPrep	Vaccinated	93.0% (119/128) (87.2%, 96.3%)	94.5% (121/128) (89.1%, 97.3%)	0.98 (0.95, 1.01)	71.0% (330/465) (66.7%, 74.9%)	70.5% (328/465) (66.2%, 74.5%)	1.01 (0.97, 1.04)
	Unvaccinated	93.5% (386/413) (90.7%, 95.5%)	93.7% (387/413) (90.9%, 95.7%)	1.00 (0.98, 1.02)	67.6% (1193/1765) (65.4%, 69.7%)	66.5% (1173/1765) (64.2%, 68.6%)	1.02 (1.00, 1.04)
SurePath	Vaccinated	91.4% (117/128) (85.3%, 95.1%)	94.5% (121/128) (89.1%, 97.3%)	0.97 (0.93, 1.00)	68.9% (314/456) (64.5%, 72.9%)	73.2% (334/456) (69.0%, 77.1%)	0.94 (0.90, 0.98)
	Unvaccinated	93.2% (383/411) (90.3%, 95.2%)	95.4% (392/411) (92.9%, 97.0%)	0.98 (0.96, 0.99)	69.7% (1205/1728) (67.5%, 71.9%)	69.6% (1203/1728) (67.4%, 71.7%)	1.00 (0.98, 1.02)

Table 47. Percent estimates and ratios of clinical sensitivities and false positive rates for the ≥CIN3 endpoints by vaccination status: Enriched population

		Clinical sensitivity			False positive rate		
Specimen Type	**Status**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**	**Alinity m HR HPV (95% CI)**	**FDA-approved test (95% CI)**	**Ratio (95% CI)**
ThinPrep	Vaccinated	96.8% (30/31) (83.8%, 99.4%)	93.5% (29/31) (79.3%, 98.2%)	1.03 (1.00, 1.12)	74.6% (419/562) (70.8%, 78.0%)	74.7% (420/562) (71.0%, 78.1%)	1.00 (0.97, 1.03)
	Unvaccinated	94.1% (127/135) (88.7%, 97.0%)	94.1% (127/135) (88.7%, 97.0%)	1.00 (0.98, 1.02)	71.0% (1450/2041) (69.0%, 73.0%)	70.1% (1431/2041) (68.1%, 72.1%)	1.01 (0.99, 1.03)
SurePath	Vaccinated	93.8% (30/32) (79.9%, 98.3%)	93.8% (30/32) (79.9%, 98.3%)	1.00 NA	72.6% (401/552) (68.8%, 76.2%)	77.0% (425/552) (73.3%, 80.3%)	0.94 (0.91, 0.98)
	Unvaccinated	94.0% (125/133) (88.6%, 96.9%)	95.5% (127/133) (90.5%, 97.9%)	0.98 (0.96, 1.00)	72.9% (1461/2004) (70.9%, 74.8%)	73.2% (1466/2004) (71.2%, 75.0%)	1.00 (0.98, 1.02)

NA: The 95% CI was not calculated as Alinity m HR HPV and FDA-approved test have the same results for every specimen.

Comparison of the Alinity m HR HPV Assay Results for Pre-Quot and Post-Quot Clinical Specimens

Additional testing was performed to compare the performance of the Alinity m HR HPV assay with ThinPrep and SurePath tested prior to cytology processing (pre-quot) or after cytology processing (post-quot). Post-quot specimens were selected using stratified random sampling. The percent agreement between pre-quot and post-quot for Alinity m HR HPV assay by clinical endpoint and genotype are summarized for each cytology processor (TP 2000, TP 5000, PrepMate/PrepStain). See **Table 48** for TP 2000 processor, **Table 49** for TP 5000 processor, and **Table 50** for PrepMate/PrepStain processor.

Table 48. Alinity m HR HPV Assay Result Pre-Quot and Post-Quot Percent Agreement: TP 2000

	Percent Agreement (n/N) (95% CI)					
Clinical Endpoint	**HPV 16**	**HPV 18**	**HPV 45**	**Other HR HPV A**	**Other HR HPV B**	**Negative**
≥ CIN2	100.0% (3/3) (43.9, 100.0)	100.0% (3/3) (43.9, 100.0)	66.7% (2[a]/3) (20.8, 93.9)	100.0% (11/11) (74.1, 100.0)	100.0% (4/4) (51.0, 100.0)	NA
< CIN2	84.6% (11/13) (57.8, 95.7)	85.7% (6/7) (48.7, 97.4)	92.3% (12/13) (66.7, 98.6)	95.5% (42/44) (84.9, 98.7)	90.7% (68/75) (82.0, 95.4)	97.4% (190/195) (94.1, 98.9)
Total	87.5% (14/16) (64.0, 96.5)	90.0% (9/10) (59.6, 98.2)	87.5% (14/16) (64.0, 96.5)	96.4% (53/55) (87.7, 99.0)	91.1% (72/79) (82.8, 95.6)	97.4% (190/195) (94.1, 98.9)

NA = not applicable. There were no Negative samples.

[a] The CIN2 case was reported as HPV 45; Other HR HPV A in the pre-quot and Other HR HPV A in post-quot.

Table 49. Alinity m HR HPV Test Result Pre-Quot and Post-Quot Percent Agreement: TP 5000

	Percent Agreement (n/N) (95% CI)					
Clinical Endpoint	**HPV 16**	**HPV 18**	**HPV 45**	**Other HR HPV A**	**Other HR HPV B**	**Negative**
≥ CIN2	100.0% (4/4) (51.0, 100.0)	NA[a]	NA[a]	100.0% (8/8) (67.6, 100.0)	100.0% (2/2) (34.2, 100.0)	NA[b]
< CIN2	92.9% (13/14) (68.5, 98.7)	90.9% (10/11) (62.3, 98.4)	89.5% (17/19) (68.6, 97.1)	85.4% (35/41) (71.6, 93.1)	94.7% (72/76) (87.2, 97.9)	100.0% (199/199) (98.1, 100.0)
Total	94.4% (17/18) (74.2, 99.0)	90.9% (10/11) (62.3, 98.4)	89.5% (17/19) (68.6, 97.1)	87.8% (43/49) (75.8, 94.3)	94.9% (74/78) (87.5, 98.0)	100.0% (199/199) (98.1, 100.0)

[a] NA = not applicable. There were no Positive samples.

[b] NA = not applicable. There were no Negative samples.

Table 50. Alinity m HR HPV Test Result Pre-Quot and Post-Quot Percent Agreement: PrepMate/PrepStain

	Percent Agreement (n/N) (95% CI)					
Clinical Endpoint	**HPV 16**	**HPV 18**	**HPV 45**	**Other HR HPV A**	**Other HR HPV B**	**Negative**
≥ CIN2	100.0% (1/1) (20.7, 100.0)	100.0% (1/1) (20.7, 100.0)	100.0% (1/1) (20.7, 100.0)	88.9% (8[a]/9) (56.5, 98.0)	100.0% (1/1) (20.7, 100.0)	NA
< CIN2	87.5% (7/8) (52.9, 97.8)	66.7% (4/6) (30.0, 90.3)	100.0% (9/9) (70.1, 100.0)	89.2% (33/37) (75.3, 95.7)	92.5% (49/53) (82.1, 97.0)	100.0% (196/196) (98.1, 100.0)
Total	88.9% (8/9) (56.5, 98.0)	71.4% (5/7) (35.9, 91.8)	100.0% (10/10) (72.2, 100.0)	89.1% (41/46) (77.0, 95.3)	92.6% (50/54) (82.4, 97.1)	100.0% (196/196) (98.1, 100.0)

NA = not applicable. There were no Negative samples.

[a] The CIN2 case was reported as HPV 45; Other HR HPV A in the pre-quot and HPV 45 in post-quot.

The agreement of the Alinity m HR HPV results prior to (pre-quot) and after (post-quot) processing on the indicated cytology processors were reported 100% for every channel at the ≥CIN2 disease endpoint with the exception of HPV 45 on the TP 2000 (66.7%) and Other HR HPV A (88.9%) on the SurePath PrepMate/PrepStain processor. As indicated above in **Table 48** and **Table 50**, the CIN2 cases were co-infected with both HPV 45 and Other HR HPV A in the pre-quot, however only one of the genotypes was positive in post-quot as indicated.

Performance with Self-Collected Vaginal Specimens

The performance of the Alinity m HR HPV assay with self-collected vaginal specimens was evaluated in a multi-center prospective clinical study. The study recruited participants ≥ 25 years of age in the US across 40 collection sites, who were undergoing routine cervical cancer screening (screening population) or were referred to colposcopy (enriched population). Each participant self-collected vaginal specimens using the simpli-COLLECT HPV Collection Kit (simpli-COLLECT) and Evalyn Brush prior to standard of care collection of a cervical specimen in ThinPrep PreservCyt Solution (ThinPrep) by the clinician. For participants in the enriched population, a colposcopy procedure was performed, and cervical histology was evaluated. simpli-COLLECT and Evalyn Brush vaginal samples were prepared for testing in the laboratory using Alinity m Specimen Buffer Kit II and Alinity m Specimen Buffer Kit III, respectively.

Clinical sensitivity was calculated for 62 study participants in the enriched population who had valid Alinity m result and histologic results of ≥ CIN2. The estimated clinical sensitivities for self-collected vaginal specimens and for clinician-collected cervical specimens, and ratio of sensitivities between the self-collected vaginal specimens and the clinician-collected cervical specimens are presented in **Table 51** for simpli-COLLECT and in **Table 52** for Evalyn Brush.

Table 51. Percent Estimates and Ratio of Clinical Sensitivity for the ≥ CIN2 Endpoint: simpli-COLLECT

Clinical Sensitivity (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
91.9% (57/62) (82.5%, 96.5%)	91.9% (57/62) (82.5%, 96.5%)	1.00 (0.93, 1.07)

Table 52. Percent Estimates and Ratio of Clinical Sensitivity for the ≥ CIN2 Endpoint: Evalyn Brush

Clinical Sensitivity (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
93.5% (58/62) (84.6%, 97.5%)	91.9% (57/62) (82.5%, 96.5%)	1.02 (0.95, 1.08)

Clinical sensitivity was calculated for 18 study participants in the enriched population who had valid Alinity m result and histologic results of ≥ CIN3. The estimated clinical sensitivities for self-collected vaginal specimens and for clinician-collected cervical specimens, and ratio of sensitivities between the self-collected vaginal specimens and the clinician-collected cervical specimens are presented in **Table 53** for simpli-COLLECT and in **Table 54** for Evalyn Brush.

Table 53. Percent Estimates and Ratio of Clinical Sensitivity for the ≥ CIN3 Endpoint: simpli-COLLECT

Clinical Sensitivity (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
88.9% (16/18) (67.2%, 96.9%)	94.4% (17/18) (74.2%, 99.0%)	0.94 (0.82, 1.00)

Table 54. Percent Estimates and Ratio of Clinical Sensitivity for the ≥ CIN3 Endpoint: Evalyn Brush

Clinical Sensitivity (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
88.9% (16/18) (67.2%, 96.9%)	94.4% (17/18) (74.2%, 99.0%)	0.94 (0.82, 1.00)

The false positive rate for simpli-COLLECT and Evalyn Brush was calculated for 724 and 713 study participants in the enriched population, respectively, who had valid Alinity m result and histologic results of < CIN2. The estimated false positive rate and ratio of false positive rate between the self-collected vaginal specimens and the clinician-collected cervical specimens are presented in **Table 55** for simpli-COLLECT and **Table 56** for Evalyn Brush.

Table 55. Percent Estimates and Ratio of False Positive Rate for < CIN2: simpli-COLLECT

False positive rate (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
74.3% (538/724) (71.0%, 77.4%)	71.5% (518/724) (68.2%, 74.7%)	1.04 (1.00, 1.08)

Table 56. Percent Estimates and Ratio of False Positive Rate for < CIN2: Evalyn Brush

False positive rate (n/N) (95% CI)		
Self-Collected Vaginal Specimens	**Clinician-Collected Cervical Specimens**	**Ratio (95% CI)**
74.2% (529/713) (70.9%, 77.3%)	71.7% (511/713) (68.3%, 74.9%)	1.04 (1.00, 1.07)

A comparison of Alinity m HR HPV assay results from self-collected vaginal specimens and clinician-collected cervical specimens was performed using paired specimens from 968 participants in the screening population and 788 participants in the enriched population for simpli-COLLECT, and from 960 participants in the screening population and 777 participants in the enriched population for Evalyn Brush. The positive percent agreements for 14 high risk HPV (ie, HR HPV Positive), HPV16/18, and 12 other HR HPV, and negative percent agreement along with 95% confidence intervals are shown in **Table 57** for simpli-COLLECT and **Table 58** for Evalyn Brush.

The percent of invalid Alinity m HR HPV test results in simpli-COLLECT specimens was 0.4% (7/1,768) with a 95% CI: 0.2% to 0.8%, and in Evalyn Brush specimens was 0.8% (14/1,756) with a 95% CI: 0.5% to 1.3%.

Table 57. Agreement Between Self-Collected Vaginal Specimens (simpli-COLLECT) and Clinician-Collected Cervical Specimens

		Clinician-Collected Cervical Specimens				
Population	**Self-Collected Vaginal Specimens**	**HR HPV Positive**	**HPV 16/18**	**12 Other**	**Negative**	**Total**
Screening	HR HPV Positive	102	14	88	93	195
	HPV 16/18	18	14	4	15	33
	12 Other	84	0	84	78	162
	Negative	4	0	4	769	773
	Total	106	14	92	862	968
	PPA (n/N) (95% CI)	96.2% (102/106) (90.7%, 98.5%)	100.0% (14/14) (78.5%, 100.0%)	91.3% (84/92) (83.8%, 95.5%)		
	NPA (n/N) (95% CI)				89.2% (769/862) (87.0%, 91.1%)	
Enriched	HR HPV Positive	540	103	437	57	597
	HPV 16/18	105	94	11	5	110
	12 Other	435	9	426	52	487
	Negative	37	2	35	154	191
	Total	577	105	472	211	788
	PPA (n/N) (95% CI)	93.6% (540/577) (91.3%, 95.3%)	89.5% (94/105) (82.2%, 94.0%)	90.3% (426/472) (87.2%, 92.6%)		
	NPA (n/N) (95% CI)				73.0% (154/211) (66.6%, 78.5%)	

Table 58. Agreement Between Self-Collected Vaginal Specimens (Evalyn Brush) and Clinician-Collected Cervical Specimens

Population	Self-Collected Vaginal Specimens	Clinician-Collected Cervical Specimens				
		HR HPV Positive	HPV 16/18	12 Other	Negative	Total
Screening	HR HPV Positive	99	13	86	70	169
	HPV 16/18	16	13	3	12	28
	12 Other	83	0	83	58	141
	Negative	5	1	4	786	791
	Total	104	14	90	856	960
	PPA (n/N) (95% CI)	95.2% (99/104) (89.2%, 97.9%)	92.9% (13/14) (68.5%, 98.7%)	92.2% (83/90) (84.8%, 96.2%)		
	NPA (n/N) (95% CI)				91.8% (786/856) (89.8%, 93.5%)	
Enriched	HR HPV Positive	536	101	435	53	589
	HPV 16/18	104	94	10	7	111
	12 Other	432	7	425	46	478
	Negative	34	4	30	154	188
	Total	570	105	465	207	777
	PPA (n/N) (95% CI)	94.0% (536/570) (91.8%, 95.7%)	89.5% (94/105) (82.2%, 94.0%)	91.4% (425/465) (88.5%, 93.6%)		
	NPA (n/N) (95% CI)				74.4% (154/207) (68.0%, 79.9%)	

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KEY TO SYMBOLS

REF	Reference Number
IVD	In Vitro Diagnostic Medical Device
LOT	Lot Number
In Vitro Test	In Vitro Test
For In Vitro Diagnostic Use	For In Vitro Diagnostic Use
AMP TRAY	AMP TRAY
ACT TRAY	ACT TRAY
UNIT	Unit
Rx ONLY	For Prescription Use Only
INFORMATION FOR USA ONLY	Information for USA Only
	Health Hazard
	Exclamation Mark
	Consult Instructions for Use or Consult Electronic Instructions for Use
	Temperature Limit
	Use By
	Contains Sufficient for <*n*> tests
	Manufacturer

TECHNICAL ASSISTANCE

For technical assistance, call Abbott Technical Services at 1-800-553-7042 (within the US) or +49-6122-580 (outside the US), or visit the Abbott website at www.molecular.abbott.

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